Wed. Sep 4, 2019

Hall A

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1015-A] Postharvest/Food Technology and Process Engineering (1)
Chair:Teodoro Espinosa-Solares(Universidad Autónoma Chapingo, Mexico), Daisuke Hamanaka(Kagoshima University, Japan)
10:15 AM - 12:00 PM Hall A (Main Hall)

[4-1015-A-01] **Influence of Maturity Stages on Postharvest Respiration Rate and Mechanical Properties of Peach Fruit**

Artemio Pérez-López[1], *Teodoro Espinosa-Solares[1], Sergio H Chávez-Franco[2], Carlos A Villaseñor-Perea[1], Luis H Hernández-Gómez[3], Consuelo Lobato-Calleros[1]（1. Universidad Autonoma Chapingo(Mexico), 2. Colegio de Posgraduados(Mexico), 3. Instituto Politecnico Nacional(Mexico)）
10:15 AM - 10:30 AM

[4-1015-A-02] **Application of the High Hydrostatic Pressure-Based Treatment as an Alternative for Food Retort Processing**

*Daisuke Hamanaka[1], Koki Morita[1], Taiga Kuhara[1], Kyohei Arimura[2], Yoshinori Kamitani[1]（1. Kagoshima Univ.(Japan), 2. Kagoshima Prefectural Osumi Food Technology Development center(Japan)）
10:30 AM - 10:45 AM

[4-1015-A-03] **Effect of High Voltage Electric Field on the Shelf Life of Mini Tomato Fruits**

*RAMNESH RAMNEEL KISHORE[1], Daisuke Hamanaka[1]（1. Kagoshima University(Japan)）
10:45 AM - 11:00 AM

[4-1015-A-04] **Impact of Low Electric Field on Physicochemical Properties and Antioxidant Activity of Persimmon (*Diospyros kaki*)**

Naruesorn Jaisue[1,2], Sutthiwal Setha[1,2], Daisuke Hamanaka[3], *Matchima Naradisorn[1,2]（1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 2. Research Group of Postharvest Technology, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 3. Faculty of Agriculture, Kagoshima University, Kagoshima, Japan(Japan)）
11:00 AM - 11:15 AM

[4-1015-A-05] **Effects of Ultrasound Pretreatments on the Texture and Colour Kinetics of Sweet Potato (Ipomea batatas) during Deep Fat Frying**

*Ayobami Olayemi Oladejo[1], Haile Ma[2], Cunshan Zhou[2]（1. University of Uyo, Uyo.(Nigeria), 2. Jiangsu University, Zhenjiang(China)）
11:15 AM - 11:30 AM

[4-1015-A-06] **Effect of Electromagnetic Field Pretreatment on Selected Vitamins and Fibre of Sweet Pepper and Fluted Pumpkin Leaf**

*Michael Mayokun ODEWOLE[1], Ayoola Patrick OLALUSI[2], Ajiboye Solomon OYERINDE[2], Olufunmilayo Sade OMOBA[3]（1. Department of Food Engineering, Faculty of Engineering and Technology, University of Ilorin, Ilorin(Nigeria), 2. Department of Agricultural and Environmental Engineering, Federal University of Technology Akure(Nigeria), 3. Department of Food Science and Technology, Federal University of Technology Akure(Nigeria)）
11:30 AM - 11:45 AM

[4-1015-A-07] **Pulsed electric fields applications in fresh meat**

*Alaa El-Din Ahmed Bekhit[1]（1. University of otago(New Zealand)）
11:45 AM - 12:00 PM

Room C

Oral Session | Food Safety

[4-1015-C] Food Safety (1)
Chair:Anthony Mutukumira(Massey University, New Zealand), siti nurjanah(Bogor Agricultural University)
10:15 AM - 12:00 PM Room C (3rd room)

[4-1015-C-01] **Surface Pasteurisation of Fresh Chicken Meat using UV-C Technology**

Arthur Jonathan Philip[1], Negah Nikanjam[1], Emilia Nowak[1], *Anthony Mutukumira[1]（1. Massey University(New Zealand)）
10:15 AM - 10:30 AM

[4-1015-C-02] **Efficient Filtering of Live *Escherichia coli* by Using 60 GHz CMOS Sensor**

*Hiroki Fukuda[1], Tetsuhito Suzuki[1], Naoshi

Kondo[1], Yuichi Ogawa[1] (1. Graduate School of Agriculture, Kyoto University(Japan))
10:30 AM - 10:45 AM

[4-1015-C-03] Safety Evaluation of Bacteriocinogenic Strains of *Pediococcus acidilactici* Isolated From Artisanal Cheeses
Luis Augusto Nero[1], Yosep Ji[2], Wilhelm Holzapfel[2], *Svetoslav Dimitrov Todorov[1] (1. Universidade Federal de Viçosa(Brazil), 2. Handong GLobal University(Korea))
10:45 AM - 11:00 AM

[4-1015-C-04] Sensitivity Comparison of Standard and real-time PCR Assay for Detection *Salmonella* Typhimurium and Enteritidis in Indonesian Chicken Carcasses
*siti nurjanah[1,2], Winiati Puji Rahayu[1,2], Ratih Dewanti-Hariyadi[1,2] (1. Department of Food Science &Technology, Bogor Agricultural University (IPB University)(Indonesia), 2. SEAFAST Center, Bogor Agricultural University (IPB University)(Indonesia))
11:00 AM - 11:15 AM

[4-1015-C-05] Development of Calculation Framework for Stochastic Prediction of Uncertainty and Variability in Survival Spore Numbers during Non-isothermal Inactivation by Second-order Monte Carlo Simulation
*Hiroki Abe[1], Kento Koyama[1], Kohei Takeoka[1], Shinya Doto[1], Shuso Kawamura[1], Shige Koseki[1] (1. Hokkaido University(Japan))
11:15 AM - 11:30 AM

[4-1015-C-06] Evaluation Growth Characteristics of Bacterial Spores Combine Treatment with High Hydrostatic Pressure and Alkaline Electrolyzed Water
*Koki Morita[1], Taiga Kuhara[1], Yoshinori Kamitani[1], Daisuke Hamanaka[1] (1. Kagoshima University faculty of agriculture(Japan))
11:30 AM - 11:45 AM

[4-1015-C-07] Impact of Mechanization Development on Women and Hired Labor Utilizations of Small-Scale Rice Farming Operations in Kampar Region, Indonesia
*UJANG PAMAN[1], Khairizal Kha, Hajry Arief Wahyudy (1. RIAU ISLAMIC UNIVERSITY(Indonesia))

11:45 AM - 12:00 PM

Room D

Oral Session | Food Quality

[4-1015-D] Food Quality (1)
Chair:Yutaka Kitamura(University of Tsukuba, Japan), Mizuki Tsuta(National Agriculture and Food Research Organization)
10:15 AM - 12:00 PM Room D (4th room)

[4-1015-D-01] Effects of Operational Conditions of Internal Combustion Furnace on rice husk Biochar and vinegar
*WEI-PUO KUO[1], YUTAKA KITAMURA[2], Yoshiyuki HARA[4], CHING-CHEN HSIEH[3], YI-HUNG LIN[3], CHEN-PIN CHEN[1] (1. Taiwan Agricultural Machinery and Biomechatromics Engineering Technology Development Association(Taiwan), 2. University of Tsukuba(Japan), 3. National Pingtung University of Science and Technology(Taiwan), 4. Hokkaido Agricultural Experiment Station(Japan))
10:15 AM - 10:30 AM

[4-1015-D-02] Assessment of Red Tomato Freshness Using Ultraviolet-induced Fluorescence Image
*Keiji Konagaya[1], Dimas Firmanda Al Riza[1], Minori Yoneda[1], Sen Nie[1], Takuya Hirata[2], Noriko Takahashi[2], Makoto Kuramoto[2], Tetsuhito Suzuki[1], Naoshi Kondo[1] (1. Kyoto Univ.(Japan), 2. Ehime Univ.(Japan))
10:30 AM - 10:45 AM

[4-1015-D-03] Thermal oxidation stability assessment of extra virgin olive oil using fluorescence and transmittance imaging system
*Vincent Rotich[1], Dimas Firmanda Al Riza[1], Ferruccio Giametta[2], Tetsuhito Suzuki[1], Yuichi Ogawa[1], Naoshi Kondo[1] (1. Kyoto University(Japan), 2. University of Molise(Italy))
10:45 AM - 11:00 AM

[4-1015-D-04] Chalkiness Index of Sake Rice "Yamada Nishiki" Using Ultraviolet-Near-Infrared Transmission
*Khokan Kumar Saha[1,2], Firmanda Al Riza Dimas[1], Yuichi Ogawa[1], Tetsuhito Suzuki[1], Naoshi Kondo[1], Takuma Sugimoto[3] (1. Lab of Bio-sensing Engineering,Graduate School of

Agriculture, Kyoto University, Kitashirakawa-Oiwakecho,Sakyo-ku ,606-8502.(Japan), 2. Department of Agricultural Engineering, Bangabandhu Sheikh Mujubur Rahman Agricultural University,Salna,Gazipur-1706.(Bangladesh), 3. Senior Researcher, Hyogo Prefectural Agriculture, Forestry and Fisheries Technology Research Center, Addo City, Hyogo Prefecture ,671-3441.(Japan))

11:00 AM - 11:15 AM

[4-1015-D-05] **Quantification of Tofu microstructure by image analysis**

*CHIANG WEN-HSIN[1], Yoshito SAITO[1], Kohei OGATA[1], Tetsuhito SUZUKI[1], Naoshi KONDO[1] （1. Graduate School of Agriculture, Kyoto University(Japan)）

11:15 AM - 11:30 AM

[4-1015-D-06] **Processing of Green Tea Paste by Micro Wet Milling and Quality Evaluation During Storage**

*Md Zohurul Islam[1], Yutaka Kitamura[1], Mito Kokawa[1], Shinya Fujii[2], Hisayuki Nakayama[2] （1. Graduate School of Life and Environmental Sciences, University of Tsukuba, Ibaraki, Tsukuba-shi, Japan(Japan), 2. Nagasaki Agricultural and Forestry Technical Development Center, Nagasaki, Japan(Japan)）

11:30 AM - 11:45 AM

[4-1015-D-07] **Quality Changes During Ripening of Mango (_Mangifera indica_ L. 'Nam Dok Mai') under Different Temperature Conditions**

*Eriko Yasunaga[1], Kohei Nakano[2], Busarakorn Mahayothee[3], Pramote Khuwijitjaru[3], Shinji Fukuda[4], Wolfram Spreer[5] （1. The University of Tokyo(Japan), 2. Gifu University(Japan), 3. Silpakorn University(Thailand), 4. Tokyo University of Agriculture and Technology(Japan), 5. Hohenheim University(Germany)）

11:45 AM - 12:00 PM

Hall A

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1330-A] **Postharvest/Food Technology and Process Engineering (2)**

Chair:Olaniyi A. Fawole(Stellenbosch University, South Africa),

nutthachai pongprasert(King Mongkut's University of Technology Thonburi, Thailand)

1:30 PM - 3:30 PM Hall A (Main Hall)

[4-1330-A-01] **Edible Coatings Control Shrivel and Maintain Quality of Nectarines during Simulated Export Conditions**

Shannon Riva[2], *Olaniyi Amos Fawole[1], Umezuruike Linus Opara[1,2] （1. Postharvest Technology Research Laboratory, South African Research Chair in Postharvest Technology, Department of Food Science, Stellenbosch University(South Africa), 2. Postharvest Technology Research Laboratory, South African Research Chair in Postharvest Technology, Department of Horticultural Science, Stellenbosch University(South Africa)）

1:30 PM - 1:45 PM

[4-1330-A-02] **Development and Characterization of Chitosan Film Incorporated with Cashew (_Anacardium Occidentale_) Leaf Extracts**

*moooksupang - Liangpanth[1,2,3] （1. Mae Fah Luang University(Thailand), 2. Thomas Karbowiak(France), 3. Wirongrong Tongdeesoontorn(Thailand)）

1:45 PM - 2:00 PM

[4-1330-A-03] **Green Synthesis of Zinc Oxide Nanoparticles from Asiatic Pennywort (_Centella asiatica_ L.) and Its Effect on the Rice Starch-Gelatin Composite Film**

*Wantida Homthawornchoo[1,2], Suttiporn Pinijsuwan[1,2], Saroat Rawdkuen[1,2] （1. School of Agro-Industry, Mae Fah Luang University(Thailand), 2. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University,(Thailand)）

2:00 PM - 2:15 PM

[4-1330-A-04] **Combination of high pressure processing and heat treatment on quality and antioxidant activity of fresh-cut persimmon**

Paweena Jarungjitaree[1], Matchima Naradisorn1[1,2], Daisuke Hamanaka [3], *Sutthiwal Setha[1,2] （1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, 57100, Thailand(Thailand), 2. Research Group of Postharvest Technology, Mae Fah Luang University, Chiang Rai, 57100,

Thailand(Thailand), 3. Faculty of Agriculture, Kagoshima University, Kagoshima, 8900065, Japan(Japan))

2:15 PM - 2:30 PM

[4-1330-A-05] **Postharvest Ethylene Application Influences Biochemical Quality of Pummelo Fruit Under Low Temperature Storage**

*Paemika Promkaew[1], Varit Srilaong[2], Satoru Kondo[1] (1. Graduate School of Horticulture, Chiba University(Japan), 2. Division of Postharvest Technology, School of Bioresources and Technology, King Mongkut's University of Technology Thonburi (Bangkhuntien)(Thailand))

2:30 PM - 2:45 PM

[4-1330-A-06] **Quality Characteristics of Thai Coconut Candy as Affected by Rice Starch-Based Film Enriched with Dragon Fruit Peel Extract**

*Wantida Homthawornchoo[1,3], Nur Fairuza Syahira Mohamad Hakimi[2,1], Saroat Rawdkuen[1,3] (1. Food Science and Technology Program, Mae Fah Luang University(Thailand), 2. Food Sciences and Technology Program, Universiti Teknologi MARA(Malaysia), 3. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University(Thailand))

2:45 PM - 3:00 PM

[4-1330-A-07] **Antifungal Packaging for Prolonging Shelf Life of Table Grapes**

*SIRIPORN LUESUWAN[1] (1. MAE FAH LUANG UNIVERSITY(Thailand))

3:00 PM - 3:15 PM

Room C

Oral Session | Food Function/Nutrition

[4-1330-C] **Functional/Wellness Foods &Nutrition (1)**

Chair:Rosires Deliza(Embrapa Food Technology, Brazil)
1:30 PM - 2:30 PM Room C (3rd room)

[4-1330-C-01] **The Influence of The Front-of-Pack Nutrition Labelling Schemes on Helping Healthier Food Choices by Consumer**

*Rosires Deliza[1], Marcela Alcantara[2], Renata

Vaqueiro Pereira[3], Gastón Ares[4] (1. Embrapa Food Technology(Brazil), 2. PDJ_CNPq/Embrapa Food Technology(Brazil), 3. Federal Rural University of Rio de Janeiro(Brazil), 4. Universidad de la Repúb lica(Uruguay))

1:30 PM - 1:45 PM

[4-1330-C-02] **Colour and Chemical Composition of Karasumi-like Chinook salmon (*O. tshawytscha*) Roe Relevant to its Quality**

*Senni Bunga[1,3], John Birch[1], Alan Carne[2], Alaa El-Din A Bekhit[1] (1. Departement of Food Science, University of Otago, New Zealand(New Zealand), 2. Department of Biochemistry, University of Otago, New Zealand(New Zealand), 3. Indonesia Endowment for Education (LPDP), Indonesia(Indonesia))

1:45 PM - 2:00 PM

[4-1330-C-03] **Effect of Inulin and *Carissa carandas* L. Supplementation on Physicochemical and Microbiological Properties of Frozen Yogurt**

*Kamonwan Manowan[1,2], Ni-orn Chomsri[1,2] (1. Agricultural Technology Research Institute, Rajamangala University of Technology Lanna(Thailand), 2. Faculty of Sciences and Agricultural Technology, Rajamangala University of Technology Lanna(Thailand))

2:00 PM - 2:15 PM

[4-1330-C-04] **Utilization of Banana Agricultural Waste: Effects of Processing Conditions on Properties of Unripe Banana (*Musa Cavendish*) Pulp and Peel Flours**

*Natthawuddhi Donlao[1,2], Asia Perin[1], Nasuha Bunyameen[1] (1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 2. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University, Thailand(Thailand))

2:15 PM - 2:30 PM

Room D

Oral Session | Food Quality

[4-1330-D] **Food Quality (2)**

Chair:Nurheni Sri Palupi(Bogor Agricultural University, Indonesia)
1:30 PM - 2:30 PM Room D (4th room)

[4-1330-D-01] Reducing Allergenicity of Soy Protein Isolate from Local Varieties of Soybean through Glycation with Lactose

*Nurheni Sri PALUPI[1,2], Endang Prangdimurti[1,2], Didah Nur Faridah[1,2], Muhammad Hasriandy Asyhari[3] （1. Department of Food Science and Technology, Faculty of Agricultural Engineering and Technology, Bogor Agricultural University(Indonesia), 2. Southeast Asian Food and Agricultural Science and Technology (SEAFAST) Center, Bogor Agricultural University(Indonesia), 3. Food Science Graduate Program, Graduate School, Bogor Agricultural University(Indonesia)）

1:30 PM - 1:45 PM

[4-1330-D-02] Nutritional Quality of Fertilized and Salted Philippine Mallard Duck (*Anas platyrhynchos L.*) Eggs Consumed in Victoria, Laguna, Philippines

*Lotis Escobin Mopera[1], Pauline Saludo[1], Floirendo Flores[1], Ma. Josie Sumague[1], Bryan Rey Oliveros[1], Wilson Tan[1] （1. Institute of Food Science and Technology, University of the Philippines Los Banos(Philippines)）

1:45 PM - 2:00 PM

[4-1330-D-03] Efficacy of 1-methylcyclopropene (1-MCP) Post-cutting Treatment on the Storage Quality of Fresh-cut 'Queen' Pineapple (Ananas comosus(L.) Merr. cv. 'Queen')

*Meryl Ancheta Bernardino[1], Katherine Anne Castillo Israel[1], Edralina Serrano[1], James Bryan Gandia[1], Wella Absulio[1] （1. University of the Philippines Los Banos(Philippines)）

2:00 PM - 2:15 PM

[4-1330-D-04] Effect of Direct and Indirect Heating On Heat Stability of Goat Milk

*Souvia Rahimah[1] （1. Universitas Padjadjaran(Indonesia)）

2:15 PM - 2:30 PM

Room C

Oral Session | Postharvest Facility

[4-1445-C] Postharvest Facility
Chair:Ahmad Al-Mallahi(Dalhousie University, Canada)
2:45 PM - 3:30 PM Room C (3rd room)

[4-1445-C-01] The Effect of Level of Fill on Nutritional Quality of Maize in an Un-aerated Clay Silos

*Mobolaji Omobowale[1], Jonathan Ogwumike[1] （1. University of Ibadan(Nigeria)）

2:45 PM - 3:00 PM

[4-1445-C-02] Pod Storage and Maturity Effects on Specialty Cacao Pulp Quality

*Jeana Cadby[1], Tetsuya Araki[1], Ian Marc Cabugsa[2] （1. University of Tokyo, Dept. Global Agricultural Sciences(Japan), 2. Ateneo de Davao University, Dept. of Chemistry(Philippines)）

3:00 PM - 3:15 PM

[4-1445-C-03] Current Status of Monitoring Post-Harvest Potato Storage Units in Atlantic Canada

*Ahmad Al-Mallahi[1] （1. Dalhousie University(Canada)）

3:15 PM - 3:30 PM

Room D

Oral Session | Postharvest Machinery

[4-1445-D] Postharvest Machinery
Chair:Yukiharu Ogawa(Chiba University, Japan)
2:45 PM - 3:15 PM Room D (4th room)

[4-1445-D-01] A Numerical Procedure for Supporting Garlic Root Trimming Machines Using Deep Learning Algorithms

*Thuyet Quoc Dang[1], Morinobu Matsuo[1,2], Takeshi Haji[1], Tetsuo Kawaide[1], Yuichi Kobayashi[1] （1. Institute of Agricultural Machinery, National Agriculture and Food Research Organization(Japan), 2. Central Region Agricultural Research Center, National Agriculture and Food Research Organization(Japan)）

2:45 PM - 3:00 PM

[4-1445-D-02] A Nondestructive Acoustic Vibration System for Apple Firmness Assessment

*Chengqiao Ding[1], Di Cui[1] （1. Zhejiang University(China)）

3:00 PM - 3:15 PM

Hall A

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1600-A] Postharvest/Food Technology and Process Engineering (3)

Chair:Lotis Escobin Mopera(University of the Philippines Los Banos, Philippines), Natthawuddhi Donlao(Mae Fah Luang University, Thailand)

4:00 PM - 6:15 PM Hall A (Main Hall)

[4-1600-A-01] Electricity Production from Xylose in Microbial Fuel Cells Started with Three Different Inoculum Sources

*Yite Liu[1], Megumi Ueda[1], Tadashi Chosa[1], Seishu Tojo[1]　(1. Tokyo University of Agriculture and Technology (Japan))

4:00 PM - 4:15 PM

[4-1600-A-02] Biodegradable Food Packaging from Cavendish Banana (*Musa acuminata*) Peduncle Fiber

Kittaporn Ngiwngam[1], Nor Jihan Jantan[2], *Wirongrong Tongdeesoontorn[1,3]　(1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai 57100 (Thailand), 2. School of Industrial Technology, Universiti Teknologi MARA, Shah Alam, Selangor 42300 (Malaysia), 3. Research Group of Innovative Food Packaging and Biomaterials, Mae Fah Luang University, Chiang Rai, 57100 (Thailand))

4:15 PM - 4:30 PM

[4-1600-A-03] Assessment of the Physical Characteristics of Maize (Zea mays) stored in different Positions within the Metallic Silos

*BABATOPE ALBERT ALABADAN[1], CALLISTUS A. OKOLO[2]　(1. Federal University, Oye Ekiti (Ikole Ekiti Campus)(Nigeria), 2. Federal University of Technology, Minna(Nigeria))

4:30 PM - 4:45 PM

[4-1600-A-04] Rice Analogue: Technology for Rice Enrichment and Food Diversification

*Lerjun Monilla Penaflor[1]　(1. Food Engineering Division, Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Banos (Philippines))

4:45 PM - 5:00 PM

[4-1600-A-05] Optimization of Process Conditions for *Batuan* [*Garcinia binucao* (Blanco) Choisy] Fruit Powder Production

*Al Kaxier Guzman Ancheta[1], Erlinda I. Dizon[2]

(1. University of the Philippines Los Banos(Philippines), 2. University of the Philippines Los Banos (Philippines))

5:00 PM - 5:15 PM

[4-1600-A-06] Effect of Processing Conditions on Bioactive Compounds and Antioxidant Activities of Tea Infusion

*Wei Qin[1], Sunantha Ketnawa[1], Florencio, Jr. Collado Reginio[1,2], Ryutaro Yamada[3], Takuya Araki[3], Yukiharu Ogawa[1]　(1. Graduate School of Horticulture, Chiba University.(Japan), 2. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Baño s(Philippines), 3. Institute of Fruit Tree and Tea Science, NARO(Japan))

5:15 PM - 5:30 PM

[4-1600-A-07] *In Vitro* Release Characteristics of Sugars and Hydrolysis of Starch During Simulated Digestion of Saba banana at Different Maturity Stages

*Florencio, Jr. Collado Reginio[1,2], Wei Qin[1], Yukiharu Ogawa[1]　(1. Graduate School of Horticulture, Chiba University(Japan), 2. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Baños(Philippines))

5:30 PM - 5:45 PM

[4-1600-A-08] *In Vitro* Examination of Starch Digestibility and Antioxidant Activities of Amaranth Grains

*Xiaoyan Xiong[1], Florencio Jr. Collado Reginio[1,2], Sukanya Thuengtung[1], Sunantha Ketnawa[1], Yukiharu Ogawa[1]　(1. Graduate School of Horticulture, Chiba University(Japan), 2. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Baño s(Philippines))

5:45 PM - 6:00 PM

[4-1600-A-09] Effects of Cell Structure Changes of Citrus Peel on the Digestibility of Intracellular Antioxidants during *in vitro* Digestion

*Yidi Cai[1], Yukiharu Ogawa[1]　(1. Graduate School of Horticulture, Chiba University(Japan))

6:00 PM - 6:15 PM

Room C

[4-1600-C] Postharvest/Food Technology and Process Engineering (4)

Chair:Kornkanok Aryusuk(King Mongkut's University of Technology Thonburi, Thailand), Itaru Sotome(University of Tokyo, Japan)
4:00 PM - 6:15 PM Room C (3rd room)

[4-1600-C-01] **Estimation of Moisture Loss of Cucumber during Storage using CFD Simulation based on Heat and Mass Transfer Model**

*Seong-Heon Kim[1], Chinatsu Nishihara[1], Fumina Tanaka[1], Fumihiko Tanaka[1] (1. Kyushu Univ.(Japan))

4:00 PM - 4:15 PM

[4-1600-C-02] **Screening (*in vitro*) The Inhibition Effect of Generally Recognized As Safe (GRAS) Substances on The Postharvest Fungal Pathogens and Its Modelling**

*Passakorn Kingwascharapong[1], Supatra Karnjanapratum[2], Fumina Tanaka[1], Fumihiko Tanaka[1] (1. Kyushu University(Japan), 2. King Mongkut's Institute of Technology Ladkrabang(Thailand))

4:15 PM - 4:30 PM

[4-1600-C-03] **Modeling The Ripening Behavior of Mature Green Tomato at Different Storage Temperatures**

*Drupadi Ciptaningtyas[1,2], Wakana Kagoshima[3], Rei Iida[1], Hitomi Umehara[1], Masafumi Johkan[1], Takeo Shiina[1] (1. Graduate School of Horticulture, Chiba University(Japan), 2. Faculty of Agro-industrial Technology, Universitas Padjadjaran(Indonesia), 3. Faculty of Horticulture, Chiba University(Japan))

4:30 PM - 4:45 PM

[4-1600-C-04] **Quality and Shelf-life Prediction of Fresh-cut 'Phulae' Pineapple by Using Image Analysis and Artificial Neural Networks**

*Rattapon Saengrayap[1], Mayura Dongsuea[1] (1. Postharvest Technology and Logistics Program, School of Agro-Industry, Mae Fah Luang University(Thailand))

4:45 PM - 5:00 PM

[4-1600-C-05] **Stationary Machine Vision Based Real-Time Estimation of Japanese Black Cattle Serum Vitamin A using Eye Fundus Color**

*SAMUEL OUMA OTIENO[1], Naoshi Kondo[1], Tateshi Fujiura[1], Yuichi Ogawa[1], Tetsuhito Suzuki[1], Katsuya Takenouchi[1], Hidetsugu Yoshioka[1], Moriyuki Fukushima[2], Takahiko Ohmae[3] (1. Graduate School of Agriculture, Kyoto University(Japan), 2. Hyogo Prefectural Hokubu Agricultural Institute(Japan), 3. Tajima Agricultural High school(Japan))

5:00 PM - 5:15 PM

[4-1600-C-06] **Segmentation of common scab lesion on intact potatoes using single near-infrared image**

*Dimas Firmanda Al Riza[1,2], Kazuya Yamamoto[3], Kazunori Ninomiya[3], Tetsuhito Suzuki[1], Yuichi Ogawa[1], Naoshi Kondo[1] (1. Laboratory of Biosensing Engineering, Graduate School of Agriculture, Kyoto University, Kitashirakawa 6068267, Kyoto(Japan), 2. Department of Agricultural Engineering, Faculty of Agricultural Technology, University of Brawijaya, Jl. Veteran 65145, Malang(Indonesia), 3. Product Planning Department, Shibuya Seiki Co., Ltd. 2200, Minamiyoshida, Matsuyama, Ehime, 791-8042(Japan))

5:15 PM - 5:30 PM

[4-1600-C-07] **Myanmar Mango Maturity Prediction Based on Skin Color Using Machine Vision System**

*RULIN CHEN[1], Dimas Firmanda Al Riza[1], Thwe Thwe Tun Naw[2], Phyu Phyu Leiyi[2], Aye Aye Thwe[2], Khin Thida Myint[1], Yuichi Ogawa[1], Tetsuhito Suzuki[1], Naoshi Kondo[1] (1. Kyoto University(Japan), 2. Yezin Agricultural University(Myanmar))

5:30 PM - 5:45 PM

[4-1600-C-08] **Measurement of Chicken Eggshell Optical Properties Using Terahertz Spectroscopy**

*Alin Khaliduzzaman[1,3], Keiji Konagaya[1], Tetsuhito Suzuki[1], Ayuko Kashimori[2], Naoshi Kondo[1], Yuichi Ogawa[1] (1. Graduate School of Agriculture, Kyoto University(Japan), 2. NABEL Co., LTd.(Japan), 3. Department of Food Engineering and Technology, Sylhet Agricultural

University(Bangladesh))
5:45 PM - 6:00 PM

[4-1600-C-09] Application of LCA (Life Cycle Assessment) Methodology in Bioethanol Production from Sugar Industry Wastewater (Molasses) – A Case Study in West Java Province, Indonesia

*Agusta Samodra Putra[1,2], Ryozo Noguchi[3], Tofael Ahamed[3] (1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Research Center for Chemistry, Indonesian Institute of Sciences(Indonesia), 3. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan))
6:00 PM - 6:15 PM

Room D

Oral Session | Others (including the category of JSAM and SASJ)

[4-1600-D] Other Categories (1)
Chair:Satoshi Yamamoto(Akita Prefectural University), Kikuhito Kawasue(University of Miyazaki)
4:00 PM - 6:15 PM Room D (4th room)

[4-1600-D-01] Applicability Of Japanese Standard About The Powered Exoskeleton To Agriculture

*Masahiro Tanaka[1], Satoru Umeno[1], Yutaka Kikuchi[1] (1. National Agriculture and Food Research Organization(Japan))
4:00 PM - 4:15 PM

[4-1600-D-02] Research on an Intelligent Robot Eye-hand System for Harvesting Pumpkin in the Outdoor Condition

*Liangliang Yang[1], Qian Wang[2], Yohei Hosino[1], Hiroki Ishikuro[1], Ying Cao[1] (1. Kitami Institute of Technology(Japan), 2. Ning Xia University(China))
4:15 PM - 4:30 PM

[4-1600-D-03] Handy Type Pig Weight Estimation System Based on Random Forest Algorithm

*Hsu Lai Wai[1], Kikuhito Kawasue[1], Khin Dagon Win[1], Kumiko Yoshida[2] (1. University of Miyazaki(Japan), 2. KOYO Plant Service(Japan))
4:30 PM - 4:45 PM

[4-1600-D-04] Plant Disease Identification using Explainable Features with Deep Convolutional Neural Network

*Harshana Habaragamuwa[1], Yu Oishi[1], Katu Takeya[1], Kenichi Tanaka[1] (1. National Agriculture and Food Research Organization(Japan))
4:45 PM - 5:00 PM

[4-1600-D-05] Sensitivity and Dynamic Analysis of Microalgae Fuel Production System Using LCA

*Riaru ISHIZAKI[1], Ryozo Noguchi[2], Agusta Samodra Putra[1], Tofael Ahamed[2], Makoto M. Watanabe[3] (1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan), 3. Algae Biomass and Energy System R&D Center, University of Tsukuba(Japan))
5:00 PM - 5:15 PM

[4-1600-D-06] An Aerial Weed Detection System for Green Onion Crops Using the You-Only-Look-Once (YOLO) Deep Learning Algorithm

Addie Ira Borja Parico[1], *Tofael Ahamed[2] (1. College of Agrobiological Resource Sciences, School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan))
5:15 PM - 5:30 PM

[4-1600-D-07] A Deep Learning and MSM Machine Learning System for Recognition of Weed Infestation in Cabbage Field Using Unmanned Aerial Vehicle

*Tofael Ahamed[1], Yan Zhang[1], Linhuan Zhang[1], Ryozo Noguchi[1] (1. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan))
5:30 PM - 5:45 PM

[4-1600-D-08] Mallard Navigation Using Unmanned Ground Vehicles, Imprinting, and Feeding

*Hirokazu Madokoro[1], Satoshi Yamamoto[1], Hanwool Woo[1], Kazuhito Sato[1] (1. Akita Prefectural University(Japan))
5:45 PM - 6:00 PM

[4-1600-D-09] Onion Bulb Counting in a Large-scale Field Using a Drone with RTK-GNSS

*Satoshi Yamamoto[1], Hirokazu Madokoro[1], Yo Nishimura[1], Yukio Yaji[1] （1. Akita Prefectural University(Japan)）
6:00 PM - 6:15 PM

Thu. Sep 5, 2019

Hall A

Oral Session | Food Safety

[5-1015-A] Food Safety (2)
Chair:Ubonrat Siripatrawan(Chulalongkorn University, Thailand)
10:15 AM - 11:30 AM Hall A (Main Hall)

[5-1015-A-04] **Cinnamon Oil Nanoemulsion as a Natural Microbial Decontaminant of Chilled Fish Flesh**

Piyanan Chuesiang[1,2], Romanee Sanguandeekul[1], *Ubonrat Siripatrawan[1,2] （1. Chulalongkorn University, Department of Food Technology, Faculty of Science(Thailand), 2. The Novel Technology for Food Packaging &Control of Shelf Life Research Group, Chulalongkorn University(Thailand)）
10:15 AM - 10:30 AM

[5-1015-A-02] **Application of Fluorescence Spectroscopy for the Classification of honey based on Geographical Origin**

*Abdullah Iqbal[1,2], Mizuki Tsuta[1] （1. Food Research Institute, National Agriculture and Food Research Organization 2-1-12 Kan-nondai, Tsukuba, Ibaraki 305-8642 Japan (Japan), 2. Dept. of Food Technology &Rural Industries, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh(Bangladesh)）
10:30 AM - 10:45 AM

[5-1015-A-03] **Preservation of sardine and scallop by high hydrostatic pressure: safety and quality aspects**

*Amauri Rosenthal[1], Rosiane Costa Bonfim[1,2], Fabiano Alves Oliveira[3], Ronoel Luiz de Oliveira Godoy[1], Carlos Adam Conte Junior[4], Eduardo Henrique Miranda Walter[1] （1. Embrapa(Brazil), 2. Federal Rural University of Rio de Janeiro(Brazil), 3. Cefet Valença(Brazil), 4. Federal Fluminense University(Brazil)）
10:45 AM - 11:00 AM

[5-1015-A-01] **Assessment of the Handling and**

Temporary Storage of Yams in Market Places in Ibadan, Oyo State, Nigeria

*Okwunna Maryjane Umego[1], Habeeb Adedotun Alabi[2], Yahaya Mijinyawa[2] （1. Federal University Oye Ekiti(Nigeria), 2. University of Ibadan(Nigeria)）
11:00 AM - 11:15 AM

[5-1015-A-05] **Responsiveness to Food Safety Emergencies in Eswatini following the Outbreak of listeriosis in South Africa**

*Tendekayi Henry Gadaga[1], Anthony N Mutukumira[2] （1. University of Eswatini(Swaziland), 2. Massey University(New Zealand)）
11:15 AM - 11:30 AM

Room C

Oral Session | Postharvest/Food Technology and Process Engineering

[5-1015-C] Postharvest/Food Technology and Process Engineering (5)
Chair:Akindele Folarin Alonge(University of Uyo, Nigeria)
10:15 AM - 11:30 AM Room C (3rd room)

[5-1015-C-01] **THE EFFECT OF DRYING METHODS ON THE QUALITY OF TIGER NUT *(Cyperus esculentus lativum)***

*Akindele Folarin ALONGE[1], Edikan Ufot GILBERT （1. University of Uyo(Nigeria)）
10:15 AM - 10:30 AM

[5-1015-C-02] **Optimization and Storage Stability Evaluation of Antioxidant Extracts From Batangas Cherry (*Terminalia microcarpa* Decne)**

*Dennis Marvin Opena Santiago[1], Shekayna Eunice Balmes Pacia[1], Jake Lloyd Cabrera Peña[1,2], Claire Solis Zubia[1], Sheba Mae Magbanua Duque[1] （1. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Banos, College, Laguna 4031 Philippines(Philippines), 2. Department of Science and Technology CALABARZON Region, Regional Science and Technology Center Complex, Jamboree Road, Timugan, Los Banos, Laguna 4030 Philippines(Philippines)）
10:30 AM - 10:45 AM

[5-1015-C-03] Effects of Pre-drying treatment and Drying-air Temperature on Moisture Ratio and Effective Moisture Diffusivity of Tomato (Nigerian Local and Foreign Varieties)

*Obafemi Ibitayo Obajemihi[1], Joshua Olanrewaju Olaoye[2], Mayowa Saheed Sanusi[1] （1. Food Engineering Department, University of Ilorin(Nigeria), 2. Agricultural and Biosystems Engineering, University of Ilorin(Nigeria)）

10:45 AM - 11:00 AM

[5-1015-C-04] Extending the Shelf-life of Upland Water Spinach (*Ipomoea aquatica*) Using Trimming, Modified Atmosphere Packaging (MAP) and Low-Temperature Storage

*Ana Mithuzela Espigol[1], Josephine Agravante[1] （1. Postharvest Horticulture Training and Research Center (PTHRC), College of Agriculture and Food Science (CAFS), University of the Philippines Los Baños (UPLB), Laguna, Philippines(Philippines)）

11:00 AM - 11:15 AM

[5-1015-C-05] Investigation of Cowpea Variety and Storage Methods on Cowpea Beetle Infestation

*VICTORIA ADA ABODENYI[1], YAHAYA MOBMI MUSA[2], ABDULLAH MUHAMMED BAKO[3] （1. Agricultural Engineering, Federal Polytechnic, Bauchi(Nigeria), 2. Federal polytechnic, Bauchi(Nigeria), 3. 1(Nigeria)）

11:15 AM - 11:30 AM

Room D

Oral Session | Others (including the category of JSAM and SASJ)

[5-1015-D] Other Categories (2)

Chair:Tri Yuliana(Universitas Padjadjaran, Indonesia)

10:15 AM - 11:30 AM Room D (4th room)

[5-1015-D-01] Screening and Enzyme Activity of Cellulose-Producing Bacteria Isolated from Kemiri Sunan (*Reutealis trisperma* (Blanco) Airy Shaw) and Empty Fruit Bunches of Palm Oil

*Tri Yuliana[1], Efri Mardawati[1], Souvia Rahimah[1], Emilda Ayu Febrianty[1], Agus Try Hartono[1] （1. Univ. Padjadjaran, Indonesia(Indonesia)）

10:15 AM - 10:30 AM

[5-1015-D-02] Development of a Cloud-based Internet of things Monitoring System for Fish Activity and Water Quality in Aquaponics

*Chien Lee[1], Yu-Jen Wang[1] （1. Department of Mechanical and Electromechanical Engineering, National Sun Yat-sen University(Taiwan)）

10:30 AM - 10:45 AM

[5-1015-D-03] EFFECT OF DIFFERENT MODES OF PLANTING AND WEEDING ON MACHINE FIELD CAPACITY AND YIELD OF A MIXED CROPPING SMALL HOLDER FARM

Folasayo Titilola Fayose[1], Adesoji Mathew Olaniyan[1], *Babatope Albert Alabadan[1], Anthony Ayodele Fajinmi[1], Kayode Ogunleye[1], Olanrewaju Omoju[1], Olufemi Aladejebi[1], Oluwaseun Ilesanmi[1] （1. Federal University Oye Ekiti(Nigeria)）

10:45 AM - 11:00 AM

[5-1015-D-04] Development of Agro-industrial Worker Trust Assessment System for Sustainable Ergonomic Program in Food Small and Medium-sized Enterprises

*Mirwan Ushada[1], Nur Achmad Sulistyo Putro[2], Titis Wijayanto[3], Fitri Trapsilawati[3], Nafis Khuriyati[1] （1. Universitas Gadjah Mada, Department of Agro-industrial Technology(Indonesia), 2. Universitas Gadjah Mada, Department of Computer Science and Electronics(Indonesia), 3. Universitas Gadjah Mada, Department of Mechanical and Industrial Engineering(Indonesia)）

11:00 AM - 11:15 AM

[5-1015-D-05] ASSESSING LAND USE TYPES IMPACT ON SOIL ORGANIC CARBON IN SOUTH WEST, NIGERIA

*OLORUNWA ERIC OMOFUNMI[1], ADESOJI MATTHEW OLANIYAN[1] （1. FEDERAL UNIVERSITY OYE-EKITI(Nigeria)）

11:15 AM - 11:30 AM

Fri. Sep 6, 2019

Hall A

Oral Session | Food Function/Nutrition

[6-1015-A] Functional/Wellness Foods &Nutrition (2)

Chair:Rungarun Sasanatayart(Mae Fah Luang University,

Thailand)
10:15 AM - 11:30 AM Hall A (Main Hall)

[6-1015-A-01] **Change of Bioactive Compounds and Bioactivities of Crisphead Lettuce during Simulated *In Vitro* Digestion**

*Sunantha Ketnawa[1], Yukiharu Ogawa[1]　(1. Graduate School of Horticulture, Chiba University(Japan))

10:15 AM - 10:30 AM

[6-1015-A-02] **Impact of Crystallinity Change During *In Vitro* Digestion on Starch Digestibility of Microwave- and Steam-Cooked Black Rice**

*Sukanya Thuengtung[1], Yoshitaka Matsushita[2], Yukiharu Ogawa[1]　(1. Graduate School of Horticulture, Chiba University(Japan), 2. Research Network and Facility Services Division, National Institute for Materials Science (NIMS)(Japan))

10:30 AM - 10:45 AM

[6-1015-A-03] **Study of Static *In Vitro* Digestion of Japanese Pickled Plums on the Change of Polyphenols and Antioxidant Activity**

*Jutalak Suwannachot[1], Sunantha Ketnawa[1], Yukiharu Ogawa[1]　(1. Chiba University(Japan))

10:45 AM - 11:00 AM

[6-1015-A-04] **Preparation of Pigment Extract Loaded Alginate Beads and Their Stability of Antioxidant Activities during *in vitro* Gastrointestinal Digestion**

*Rungarun Sasanatayart[1], Sutthiwal Setha[1]　(1. School of Agro-Industry, Mae Fah Luang University(Thailand))

11:00 AM - 11:15 AM

[6-1015-A-05] **Stability of Plant Pigments and Antioxidant Activities in Juice Model during Processing and *in vitro* Gastrointestinal Digestion**

*Titikan Liangpanth[1], Rungarun Sasanatayart[1]　(1. School of Agro-Industry, Mae Fah Luang University(Thailand))

11:15 AM - 11:30 AM

Room C

Oral Session | Postharvest/Food Technology and Process Engineering

[6-1015-C] **Postharvest/Food Technology and Process Engineering (6)**

Chair:Xujun Ye(Hirosaki University, Japan)

10:15 AM - 11:30 AM Room C (3rd room)

[6-1015-C-01] **Spatially Resolved Interactance Spectroscopy to Estimate Degree of Red Coloration in Red-fleshed Apple Cultivar 'Kurenai-no-Yume'**

*Xujun Ye[1], Sou Takada[1], Shuhuai Zhang[1]　(1. Hirosaki University(Japan))

10:15 AM - 10:30 AM

[6-1015-C-02] **Use of hyperspectral imaging to separate cultivars and evaluate the internal quality of nectarines**

Sandra Munera[1], Prieto Andres[1], Nuria Aleixos[2], Sergio Cubero[1], *Jose Blasco[1]　(1. Centro de Agroingeniería. Instituto Valenciano de Investigaciones Agrarias (IVIA). Ctra. Moncada-Náquera Km 4.5, 46113, Moncada, Valencia(Spain), 2. Departamento de Ingeniería Gráfica. Universitat Politècnica de València. Camino de Vera, s/n, 46022 Valencia(Spain))

10:30 AM - 10:45 AM

[6-1015-C-03] **Evaluating the Performance of Unmanned Crop Sensing Robot for Rice**

*Dhirendranath Singh[1], Shigeru Ichiura[1], Mitsuhiko Katahira[2,1]　(1. United Graduate School of Agriculture , Iwate University(Japan), 2. Faculty of Agriculture, Yamagata University(Japan))

10:45 AM - 11:00 AM

[6-1015-C-04] **Application of Non-destructive Determination of Rice Amylose Content at Grain Elevators**

*Edenio Olivares Diaz[1], Shuso Kawamura[1], Miki Matsuo[1], Toru Nagata[2], Shigenobu Koseki[1]　(1. Hokkaido University(Japan), 2. Hokkaido Research Organization Central Agricultural Experiment Station(Japan))

11:00 AM - 11:15 AM

[6-1015-C-05] **Cow Milk Progesterone Concentration Determination during Milking Using Near-infrared Spectroscopy**

*Patricia Nneka Iweka[1], Shuso Kawamura[1], Tomohiro Mitani[2], Takashi Kawaguchi[3], Shigenobu Koseki[1]　(1. Hokkaido Univ.(Japan), 2. Field Sc. Center(Japan), 3. Orion Mach.(Japan))

11:15 AM - 11:30 AM

Room D

[6-1015-D] **Other Categories (3)**
Chair:Takahiro Orikasa(Iwate University, Japan)
10:15 AM - 11:30 AM Room D (4th room)

[6-1015-D-01] **Field Representation and Path Planning for Robot Tractors**

*Hao Wang[1], Noboru Noguchi[1]（1. Hokkaido University(Japan)）

10:15 AM - 10:30 AM

[6-1015-D-02] **Driving Force Control for Suppression of Tractor's Dynamic Pitching Angle**

*Yuya Aoyagi[1], Masami Matsui[2]（1. Tokyo University of Agriculture and Technology (Japan), 2. Utsunomiya University(Japan)）

10:30 AM - 10:45 AM

[6-1015-D-03] **Development of a Smart Spraying System For Weeds On Rice Fields**

*Thanh Tinh Nguyen[1], Ricardo Ospina[2], Noboru Noguchi[2]（1. Hokkaido University, Graduate School of Agriculture(Japan), 2. Hokkaido University, Research Faculty of Agriculture(Japan)）

10:45 AM - 11:00 AM

[6-1015-D-04] **Deep Learning and Multiple Sensors Data Acquisition System for Real-time Decision Analysis in Agriculture Using Unmanned Aerial Vehicle**

*Yunyan Xie[1], Ryozo Noguchi[2], Tofael Ahamed[2]（1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan)）

11:00 AM - 11:15 AM

[6-1015-D-05] **Autonomous Navigation and Obstacle Avoidance for a Robotic Mower using Machine Vision**

*Kosuke Inoue[1]（1. The University of Tokyo(Japan)）

11:15 AM - 11:30 AM

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1015-A] Postharvest/Food Technology and Process Engineering (1)

Chair:Teodoro Espinosa-Solares(Universidad Autónoma Chapingo, Mexico), Daisuke Hamanaka(Kagoshima University, Japan)
Wed. Sep 4, 2019 10:15 AM - 12:00 PM Hall A (Main Hall)

[4-1015-A-01] **Influence of Maturity Stages on Postharvest Respiration Rate and Mechanical Properties of Peach Fruit**

Artemio Pérez-López[1], *Teodoro Espinosa-Solares[1], Sergio H Chávez-Franco[2], Carlos A Villaseñor-Perea[1], Luis H Hernández-Gómez[3], Consuelo Lobato-Calleros[1] （1. Universidad Autonoma Chapingo(Mexico), 2. Colegio de Posgraduados(Mexico), 3. Instituto Politecnico Nacional(Mexico)）
10:15 AM - 10:30 AM

[4-1015-A-02] **Application of the High Hydrostatic Pressure-Based Treatment as an Alternative for Food Retort Processing**

*Daisuke Hamanaka[1], Koki Morita[1], Taiga Kuhara[1], Kyohei Arimura[2], Yoshinori Kamitani[1] （1. Kagoshima Univ.(Japan), 2. Kagoshima Prefectural Osumi Food Technology Development center(Japan)）
10:30 AM - 10:45 AM

[4-1015-A-03] **Effect of High Voltage Electric Field on the Shelf Life of Mini Tomato Fruits**

*RAMNESH RAMNEEL KISHORE[1], Daisuke Hamanaka[1] （1. Kagoshima University(Japan)）
10:45 AM - 11:00 AM

[4-1015-A-04] **Impact of Low Electric Field on Physicochemical Properties and Antioxidant Activity of Persimmon (*Diospyros kaki*)**

Naruesorn Jaisue[1,2], Sutthiwal Setha[1,2], Daisuke Hamanaka[3], *Matchima Naradisorn[1,2] （1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 2. Research Group of Postharvest Technology, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 3. Faculty of Agriculture, Kagoshima University, Kagoshima, Japan(Japan)）
11:00 AM - 11:15 AM

[4-1015-A-05] **Effects of Ultrasound Pretreatments on the Texture and Colour Kinetics of Sweet Potato (Ipomea batatas) during Deep Fat Frying**

*Ayobami Olayemi Oladejo[1], Haile Ma[2], Cunshan Zhou[2] （1. University of Uyo, Uyo.(Nigeria), 2. Jiangsu University, Zhenjiang(China)）
11:15 AM - 11:30 AM

[4-1015-A-06] **Effect of Electromagnetic Field Pretreatment on Selected Vitamins and Fibre of Sweet Pepper and Fluted Pumpkin Leaf**

*Michael Mayokun ODEWOLE[1], Ayoola Patrick OLALUSI[2], Ajiboye Solomon OYERINDE[2], Olufunmilayo Sade OMOBA[3] （1. Department of Food Engineering, Faculty of Engineering and Technology, University of Ilorin, Ilorin(Nigeria), 2. Department of Agricultural and Environmental Engineering, Federal University of Technology Akure(Nigeria), 3. Department of Food Science and Technology, Federal University of Technology Akure(Nigeria)）
11:30 AM - 11:45 AM

[4-1015-A-07] Pulsed electric fields applications in fresh meat

*Alaa El-Din Ahmed Bekhit[1] （1. University of otago(New Zealand)）

11:45 AM - 12:00 PM

10:15 AM - 10:30 AM （Wed. Sep 4, 2019 10:15 AM - 12:00 PM Hall A)

[4-1015-A-01] Influence of Maturity Stages on Postharvest Respiration Rate and Mechanical Properties of Peach Fruit

Artemio Pérez-López[1], *Teodoro Espinosa-Solares[1], Sergio H Chávez-Franco[2], Carlos A Villaseñor-Perea[1], Luis H Hernández-Gómez[3], Consuelo Lobato-Calleros[1] （1. Universidad Autonoma Chapingo(Mexico), 2. Colegio de Posgraduados(Mexico), 3. Instituto Politecnico Nacional(Mexico)）
Keywords: physical properties, maturity stage, respiration rate, compression load, anisotropy, apparent elasticity modulus

The objective of this work is to elucidate the influence of maturity stage of peach fruit cv. Diamante on the respiration rate and mechanical properties (compression load, strain and apparent elasticity modulus during storage at room temperature. Three maturity stages (green, middle yellow and yellow) were selected for the study. The highest respiration rate was observed for middle yellow fruits. For compressive load, mechanical properties of peach fruit tissue showed anisotropic behavior in the orientation tangential or radial directions. Peach maturity stage strongly influenced the mechanical properties of peach. The physical properties sphericity, density, bulk density, porosity, and packing coefficient of the fruit harvested at physiological maturity stage were also evaluated.

10:30 AM - 10:45 AM （Wed. Sep 4, 2019 10:15 AM - 12:00 PM Hall A)

[4-1015-A-02] Application of the High Hydrostatic Pressure-Based Treatment as an Alternative for Food Retort Processing

*Daisuke Hamanaka[1], Koki Morita[1], Taiga Kuhara[1], Kyohei Arimura[2], Yoshinori Kamitani[1] （1. Kagoshima Univ.(Japan), 2. Kagoshima Prefectural Osumi Food Technology Development center(Japan)）
Keywords: bacterial spores, heat resistance, hydrostatic pressure, shelf life

In order to extend the shelf-life of various foods, it is seriously essential to install some appropriate sterilizing technologies. In particular, various foods expecting to be stored for a long period are required to be in almost microbial-eliminated during processing. For satisfying such requirement, typical retort treatment with 121℃ for more than 5 min has been performed in the food industry. The main microbes targeted in retort processing are spores formed by some Gram positive bacteria. Bacterial spores are known to be extremely resistant to various stresses physically and chemically as compared to vegetative cells. Most of the spores are of soil origin, which means the fact that almost all agricultural products can be regarded as being contaminated with bacterial spores. Therefore, in order to preserve processed foods including agricultural commodities for a long period of time, it is required to introduce a technique capable of effectively inactivating bacterial spores. However, the current major problem is that no appropriate method other than retort treatment can be found. So far, we have investigated the effective treatment for the reduction of heat resistance of bacterial spores mainly based on high hydrostatic pressure(HHP). In particular, it has been shown that the reduction efficiency is accelerated by the combined use with alkaline electrolyzed water(AlEW), but, any experimental trials for actual processed foods has not been conducted yet. In this study, we investigated the application possibility of HHP-based treatment for extending shelf-life of some processed foods inoculated with typical bacterial spores. *B. subtilis*, *B. cereus* and *C. sporogenes* were used as test bacteria. After aerobically and anaerobically grown with appropriate media for *Bacillus* and *Clostridium*, respectively, spores were formed on the plate medium culture. After preparation of spore suspension with its spectrophotometric absorbance at 500 ㎚ adjusted to 1 according to the conventional

methodology, the same amount of each spore suspension was mixed to prepare a spore cocktail, then inoculated to material of the processed foods. In this experiment, hamburg and potato salad were used as test foods. The ingredients used in hamburg were minced pork, diced onion, and that in potato salad were potato and mayonnaise, respectively. No additional ingredients such as spices were used. All materials used were obtained from supermarket neighboring Kagoshima University. The material inoculated with spores was sealed in a plastic film bag with triplicate volume of sterile distilled water or AlEW, and subjected to HHP treatment. The treatment conditions were 80 MPa, 50℃ for 30 min, and the samples after treatment were prepared with hamburg and potato salad by a general cooking method. The cooked sample were heat-treated in water bath at 80℃ for 15 min, and then a storage test was conducted at 30℃. For both hamburg and potato salad, the increases in viable count of food samples treated by single HHP and HHP combined with AlEW were slower comparing with that of the untreated sample during storage. More than 2 days in storage, 4-5 logs differences were obtained between the treated and untreated sample. At the same time, gas production in sealed package of treated sample was not observed. Since the storage test was in an unaerobic state, bacterial growth was considered to be dominated by *Clostridium*, but it is considered that *Bacillus* could be also effectively inactivated considering our previous unpublished data. From the above, it was shown that the treatment based on HHP is as effective as the *in vitro* study in reducing the heat resistance of bacterial spore, and it may be possible to replace retort treatment.

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1015-A] Postharvest/Food Technology and Process Engineering (1)

Wed. Sep 4, 2019 10:15 AM - 12:00 PM Hall A (Main Hall)

[4-1015-A-03] Effect of High Voltage Electric Field on the Shelf Life of Mini Tomato Fruits

*RAMNESH RAMNEEL KISHORE[1], Daisuke Hamanaka[1] （1. Kagoshima University(Japan)）
Keywords: High voltage electric field, climacteric fruits, respiration rate, Tomato

High voltage electric field (HVEF) which is a non thermal food preservation technique that processes food without leaving any chemical residues and radiation is one of the emerging and a new area of research in the field of food preservation. Recent research indicates the use of HVEF on agricultural commodities reduces respiration rates of climacteric fruits, inhibit enzymatic and microbial activity which leads to extending the shelf life of fruits and vegetables. A major advantage of using high voltage food preservation technique is that it consumes low energy and prohibits the use of any chemical preservatives. The main objective of this research is to investigate the effects of HVEF on the shelf extension of mini tomato fruits. Monitoring of physiological responses and quality evaluation were performed during the storage test period of 3 weeks. The mini tomato fruit samples were obtained from a local distributor in Kagoshima and the samples were carefully graded for optimum quality to be used in this experiment. The total duration of sample analysis was 3 weeks (21 days). In the first two weeks the samples were treated to continuous electric field treatment; this was the test to study continuous effect of electric field on sample quality. On the 3rd week; the electric field was shut off and samples were analyzed for further 1 week to test the shelf life storage stability of tomato fruits post EF treatment. For EF and control treatment the total frequency of monitoring or analysis was 7 times; which is a representation of different number of day intervals over the 2 weeks period. For shelf life storage testing period the samples were analyzed over a period of 1 week. Two distinctive type of EF setup was used to study the quality attributes and respiration rate respectively. For measurement of quality attributes, the samples were placed inside a large electric field incubator (High voltage 7kV A/C- power supply (N- TeFe II) at 60Hz) for EF treatment and for control experiment another similar type incubator was used but without EF treatment. The treatment temperature was 1^0C and relative humidity was approximately 95%. The quality attributes evaluated in this study were PLM (Physiological loss of mass), TSS (total soluble solid), Brix, hardness and color properties. For respiration rate measurement (CO_2) production, 2kV aluminum parallel plate configuration electric field setup was used. The HVEF system consisted of a power source with an output voltage of 2kV (NF Corporation Japan, Model: EC750SA) with A/C- power supply (sine waveform) at a frequency of 60Hz. The samples were placed into acrylic desiccator and sealed off with an air tight lid. One end of the desiccator was connected to the air pump via flexible tube while the other end was connected to much smaller desiccator that housed the CO_2 sensor. For control, similar setup was used but without EF plates in the desiccator. The entire system was setup inside the incubator with controlled temperature and RH. Four incubators with EF setup were used, each with a corresponding temperature of 0, 1, 3, 5^0C respectively. The CO_2 production of the fruits were assessed at 10 minute intervals for 2 hours. For post EF treatment analysis, the samples were transferred inside perforated bag with gas septum attached and the bags were sealed and further stored for 1 week to study shelf life stability. The gas concentration was analyzed using a gas chromatography machine. The experiments were performed in triplicates. Analysis of the PLM indicated that in the two weeks of continuous treatment, the electric field treated samples had suppressed weight loss on D1, D2 and D3 respectively, whereas for D7 and D10 samples; the control treatment showed more reduction in weight loss. During shelf life storage period, the PLM of EF samples was significantly lower than the control treatment. On day 7, maximum weight loss was seen, however the weight

loss of EF sample was still lower compared to control 0.60 and 0.87% respectively. The total soluble solid content of HVEF treated samples were higher in concentration compared to the control in the 2 weeks of continuous treatment. Post EF treatment, the EF samples still constituted to higher Brix content overall. On the day 7 of shelf life storage the average TSS content between EF and control sample were 7.06 and 6.52 Brix respectively. The fruit firmness during 2 weeks of continuous treatment indicated that on day 4 and 7, the EF samples were a lot slightly firmer with 0.84N and 0.83N difference between EF and control sample respectively. Post EF treatment, the EF treated sample were consistently firmer throughout the 1 week storage period with an average hardness of 6.7N for EF sample and 6.5N for control.

Effect of High Voltage Electric Field on the Shelf Life of Mini Tomato Fruits.

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ABSTRACT

In this experiment two distinct type of electric field setup was used to study quality attribute and respiration rate of tomato fruit (*Solanum lycopersicum*) during 2 weeks of continuous electric field treatment and 1 week of post electric field shelf life storage testing. The main focus of this experiment is to study the effects of electric field on the shelf life stability of tomato fruit. For studying quality attribute (PLM, Brix, color & hardness), a 7kV alternating current electric field setup was used. The results of samples treated with electric field had an overall suppressed weight loss, higher concentration of ^{o}Brix and firmer texture during 1 week of shelf life testing period compared to control. The Hunters color value showed no significant difference between electric field and control samples. For measurement of respiration rate (CO_2 yield), a 2kV aluminum parallel plate setup at four temperature levels of 0, 1, 3 & 5°C was used in this experiment. During 2 weeks of continuous electric field treatment samples exposed to 5°C had significantly lower CO_2 $_{yield}$ overall. For post electric field shelf life testing, samples stored under 3°C had least CO_2 production and comparatively electric field treated sample had suppressed CO_2 gas concentration than the control sample.

Keywords:
High voltage electric field
Mini tomato
Respiration rate
Shelf life
Post electric field
CO_2 yield

1. INTRODUCTION

Food preservation processes are important in extending the shelf life of foods. Fruits and vegetables due to their high metabolic activity and water content, tend to deteriorate in their quality rapidly once they are harvested. The reason as explained by (Tucker, 1993) is due to its metabolic activity that continues to occur even after harvest, thus making most fruits highly perishable commodities. This change is irreversible thus to maintain optimum quality and safety of the commodity effective preservation measures are paramount. Many factors are responsible in spoilage of agricultural commodities after harvest. The major factors that affect food spoilage include water activity, exposure to light and oxygen, pH, temperature, microbial and chemical factors such as enzymatic activities. As stated by (Anaparti, 2019) there are several factors that act together and their collective impact results in the complete decay of the food commodity. Therefore, the need for new post-harvest preservation technologies are very important for the extension of shelf life without significant loss in quality, freshness and consumer demands.

The existing post-harvest treatment facilities offers to be effective in fruit and vegetable preserving; however each kind of treatment facility has some limitations. Modified Atmospheric Packaging (MAP), for instance uses N_2/CO_2 stable gases and at low temperature environment for fruit and vegetable preservation storage but a study by (El-Kazzaz, M.K., Sommer, N.F. & Fortlage, R.J., 1983) reported that off-odors were produced when strawberries were kept under higher CO_2 atmosphere for more than 4 days as a result of anaerobic respiration. Elevated CO_2 (10% to 40%) was found to

degrade internal color and caused a decrease in anthocyanin content of the internal tissue of strawberry (Gil, 1997). Modified atmosphere packaging can help to extend the shelf-life of strawberries but the results can be variable (Shamaila, 1992). Another commonly used preservation technique is low temperature cold storage usually above 0^oC. The conventional cold storage temperature is 5^0C or below that; but its effects can be seen as chilling injury to fruits and vegetables. Chilling injury is simply damage to fruits and vegetables caused by temperature above the freezing point (0^0C) (Grant, 2019). At these temperature, the tissues weaken because they are not able to carry out normal metabolic process. Various physiological and biochemical alterations occur in the sensitive species in response to low temperature exposure. These alterations lead to the development of a variety of chilling injury symptoms, such as surface pitting, discoloration, internal breakdown, failure to ripen, growth inhibition, wilting, loss of flavor, and decay. Further research on new techniques and technologies on improved post-harvest preservation are therefore important.

High voltage electric field (HVEF) which is a non thermal food preservation technique that processes food without leaving any chemical residues and radiation is one of the emerging and a new area of research in the field of food preservation. Recent research indicates the use of HVEF on agricultural commodities reduces respiration rates of climacteric fruits, inhibit enzymatic and microbial activity which leads to extending the shelf life of fruits and vegetables. A major advantage of using high voltage food preservation technique is that it consumes low energy and prohibits the use of any chemical preservatives. Hence use of high voltage electric field (HVEF) is one the ways that tend retain quality and freshness of fruits and vegetables without any significant loss of quality attributes. As described by (Muthukumaran, 2009) in their paper that a major advantage of this technique is that it does not cause any significant change in food temperature during processing thus increasing this technology's capability to be applied to any temperature sensitive food matrix example fruits and vegetables

Previous researches carried out in this field suggest that the use of HVEF increases the shelf life of fruits and vegetables without undesirable heat, chemical and microbial effects. According to research by (Bajgai, et al., 2005) showed that upon treatment of Wase Satsuma mandarin fruits with alternating HVEF, the results showed that the fruits inhibited the degradation of the chlorophyll and flavonoids of the peels hence retaining color and flavor.

Furthermore, Emblic fruit (*Phyllanthus emblical L*) a fruit found in South Asia popular for its high vitamin C has also been studied under HVEF. According to the article, the fruits were treated by alternating current (AC) direct current (DC) with high voltage electric field of 430kV/m for 2 hours to study the physiological loss of mass (PLM). The parameters tested were rotting, color change and Vit. C content during storage at 4, 20 and 35 ^0C in open and closed polyethylene pouches respectively. Based on the findings of this experiment, the results showed that alternating current (HVEF) is found to have an extended shelf life of emblic fruit. The AC HVEF treated emblic fruits when tested for their hunter color values, rotting and Vit. C content showed that HVEF treated samples were much better in quality and freshness compared with the untreated samples (Bajgai, et al., 2005).

The main aim of this study is to evaluate the effects of high voltage electric field on the on the shelf extension of mini tomato fruits. Monitoring of physiological responses and quality evaluation were performed during storage test over a 3 week period.

2. MATERIALS AND METHODS

2.1 Sample preparation
 Tomato fruit (*Solanum lycopersicum*) was procured form a local distributor in Kagoshima prefecture. The sample were carefully checked and graded for optimum quality, free of any visual defects like bruises and cuts. Samples which showed signs of molds were discarded.
2.1.1 AC- HVEF experimental set up & sample treatment plan

Fig. 1 HVEF setup used in measurement of quality attributes. 1; High voltage AC power supply, 2; conducting electrode, 3; incubator, 4; storage shelf, 5; temperature monitor

For measurement of quality attributes, the samples were placed inside a large electric field incubator (High voltage 7kV A/C- power supply (N- TeFe II) at 60Hz) (Figure 1) for EF treatment; and for control experiment another similar type incubator was used but without EF treatment. The treatment temperature was 1^0C and relative humidity was approximately 95%. The quality attributes evaluated in this study were PLM (Physiological loss of mass), TSS (total soluble solid), hardness and color properties. The total duration of sample analysis was 3 weeks (21 days). In the first two weeks the samples were treated to continuous electric field treatment; this was the test to study continuous effect of electric field on sample quality. On the 3rd week; the electric field was shut off and samples were analyzed for further 1 week to test the shelf life storage stability of tomato fruits post EF treatment. For EF and control treatment the total frequency of monitoring or analysis was 7 times; which is a representation of different number of day intervals (day 1,2,3,4,7,10 & 14) over the 2 weeks period. For shelf life storage testing period the samples were analyzed over a period of 1 week. The monitoring was 5 times (day 1,2,3,4 &7) in 1 week period.

2.1.2 Measurement of Physiological loss of mass (PLM)
In this experiment the cumulative loss of mass due to moisture loss of continuous electric field treated samples, post electric field shelf life test samples and control samples of tomato fruits were measured by weighing using a balance (Mettler Toledo PB3002-S, Switzerland ±0.01g). A total of 5 fruit samples were used for EF and control measurements respectively. The difference of weight for this five samples were then determined and their mean and standard deviation were calculated.
2.1.3 Measurement of Hunters Color value and total color difference
The Hunters color values units using the CIELAB ((Commission Internationale de l'Eclairage) color parameters expressed as *L* (lightness), *a* (green chromacity), and *b* (yellow chromacity) of continuous electric field treated samples, post electric field shelf life test samples and control samples of tomato fruits were measured on day 0 and on each designated day interval over 21 days of experiment period using a color meter (Minolta Chroma Meter Model CR-10 Plus (Minolta Tokyo, Japan)). For consistent reading, the fruit samples were marked at three place along the top portion of the fruit and the color reading were taken from middle part of the fruit which was just below the marked spots. Using equation 1 and CIELAB coordinate system of *L, a, b color* reading values were used to calculate the total color difference (▲*E*) (Chen, 2013). The Hunters color values reading of five samples were taken. The mean and SD of *L, a, b* values of fruit samples were determined.

$$\Delta E = \left[\left(L^* - L_0^*\right)^2 + \left(a^* - a_0^*\right)^2 + \left(b^* - b_0^*\right)^2 \right]^{1/2} \quad \text{(Eq 1)}$$

Where L_o, a_o, b_o are the initial values of fresh tomato samples on day 0 and *L, a, b* are final values
2.1.4 Measurement of Total Soluble Sugar Concentration or Brix
The total Brix content of the of continuous electric field treated samples, post electric field shelf life test samples and control samples of tomato fruits were determined by carefully slicing the fruit into

half. 1-3 drops of the tomato juice was squeezed onto the prism of the Brix meter and the readings were taken. Brix meter (Hybrid PAL-BXACID F5) model was used. The Brix readings of triplicate samples were taken. Mean and SD were then calculated.

2.1.5 Measurement of Fruit Firmness

For measurement of fruit hardness, a Rheoner II (Creep Meter PE2 330 SC) texture analyzer was used. A P79 (2mm) cylindrical needle probe was used to penetrate the fruit which was cut from the middle along the equatorial region and carefully the cut side was placed flat on base stage of the meter. The amount of force required to penetrate the fruit was recorded in Newton (N). The fruit hardness data of triplicate samples were taken and their mean and SD were calculated.

2.1.6 Measurement of Respiration rate (CO2 yield)
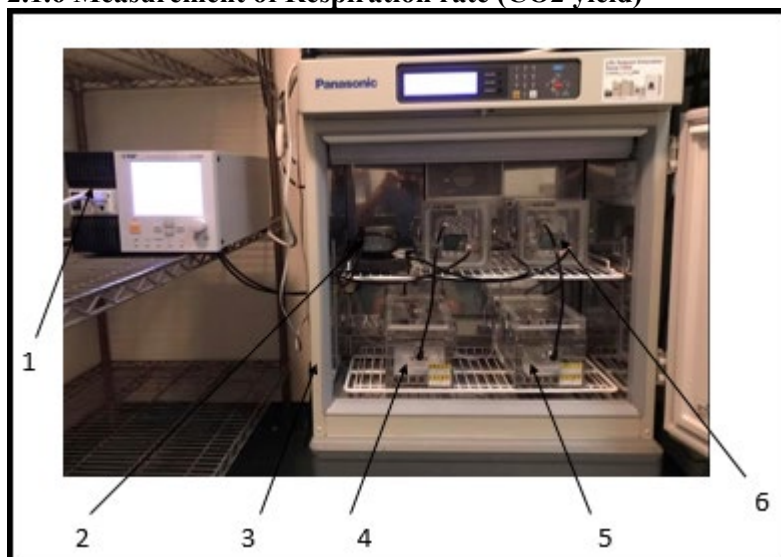
Fig. 2 HVEF setup used in measurement of respiration rate; 1; 2kV AC current power generator, 2; air pump, 3; Incubator, 4; acrylic desiccator for control treatment, 5; acrylic desiccator with parallel plate aluminum electrode, 6; smaller acrylic desiccator with CO_2 sensor.

For respiration rate measurement (CO_2) production, 2kV aluminum parallel plate configuration electric field setup was used. The HVEF system consisted of a power source with an output voltage of 2kV (NF Corporation Japan, Model: EC750SA) with A/C- power supply (sine wave) at a frequency of 60Hz. The samples were placed into acrylic desiccator and sealed off with an air tight lid. One end of the desiccator was connected to the air pump via flexible tube while the other end was connected to much smaller desiccator that housed the CO_2 sensor. For control, similar setup was used but without EF plates in the desiccator. The entire system was setup inside the incubator with controlled temperature and RH. Four incubators with EF setup were used, each with a corresponding temperature of 0, 1, 3, 5^0C respectively. The CO_2 production of the tomato fruits were measured as total gas accumulated on each day of analysis.

On day 14 the electric field was shut off. For post EF shelf life storage testing treatment, the samples were transferred inside perforated bag and the gas septum was attached on the upper side and the bags were sealed and further stored for 1 week at the same corresponding temperature of 0, 1, 3, 5^oC to study shelf life stability. The CO_2 gas production was analyzed using a gas chromatography machine (GC-4000 PLUS). The experiments were performed in triplicates. The mean and SD were calculated.

3. RESULTS AND DISCUSSION

3.1.1 Effect of HVEF on Physiological loss of mass of tomato fruit

Results of analysis of physiological loss of mass (PLM) of tomato fruits is shown in Table 1. The percent loss of mass as moisture loss was in a range of 0.21% to 2.12% for continuous electric filed treated sample of 2 weeks of treatment while for control treatment it was in the range of 0.31% to 2.08%. As shown by the Figure 3 the control sample had slightly higher weight loss on day 1 to day 3, however from day 4 onwards it was observed that the continuous electric field treated sample had a higher weight loss. For post electric field shelf life testing of 1 week, the results showed samples

which were treated with EF had an overall suppressed weight loss in range of 0.13- 0.60% after a week while the control samples were slightly higher in range of 0.20 – 0.80%. For both treatment types the weight loss was less than 1%. This findings suggest continuous electric treatment had a positive effect towards suppressing the weight loss of tomato fruit during shelf life test period.

Table1
Continuous effects of electric field treatment on Physiological loss of mass (of tomato fruits) treated for 2 weeks & Post EF shelf life storage test of 1 week at 1ºC and 95% RH

Treatment Interval (days)	HVEF samples (%)	Control sample (%)	Treatment Interval (days)	HVEF samples (%)	Control sample (%)
	Continuous Electric Field treatment			Post EF shelf life Test	
1	0.21 ± 0.01	0.31 ± 0.10	1	0.13 ± 0.05	0.20 ± 0.04
2	0.32 ± 0.02	0.46 ± 0.10	2	0.25 ± 0.05	0.31 ± 0.04
3	0.45 ± 0.07	0.55 ± 0.09	3	0.36 ± 0.05	0.44 ± 0.03
4	0.67 ± 0.11	0.55 ± 0.09	4	0.49 ± 0.05	0.64 ± 0.04
7	1.08 ± 0.13	0.66 ± 0.26	7	0.60 ± 0.05	0.88 ± 0.09
10	1.51 ± 0.20	1.07 ± 0.03			
14	2.12 ± 0.34	2.08 ± 0.10			
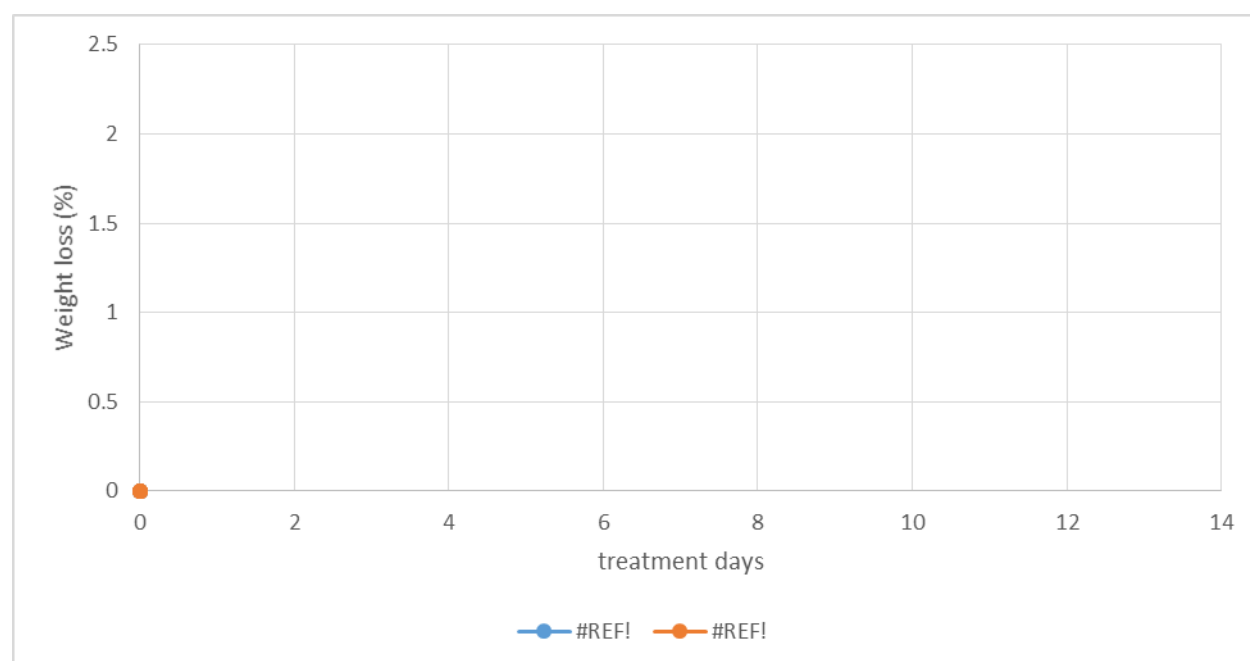
Fig 3. Physiological loss of mass of HVEF and Control samples after 14 days (1^{0}C; 95% RH)

3.1.2 Effect of HVEF on Hunters color value and total color change
The results from the Hunters color value of continuous electric filed treated samples and post electric field shelf life testing sample data are shown in the Table 2 & 3. For continuous EF treatment and control samples very little difference were evident between the initial and final color value (*L, a, b*) readings from day 0 to day 14. The results showed that difference between control sample and EF treated samples were very insignificant. The total color difference (▲E) was in range of 1.46 to 2.80 for EF samples and 0.74 to 2.97 for control samples. This value indicated fairly small difference. Except for day 10, where (▲E) value was maximum for both EF sample (5.72) and control (5.12) respectively. This increase in ▲E could have been result of some chemical or enzymatic activities that might have triggered. For post EF shelf life testing period, similar results were observed. There was very little difference between post EF treated samples and control samples except for day 7. On day 7 the total color difference was 4.61 for EF sample and 4.10 of control sample. Values in this range indicated perceptible difference between the color values.

Table2

Effects of HVEF on the Hunters color value of tomato fruit during 2 weeks continuous treatment

treatment interval (days)	HVEF sample				Control sample			
	L	a	b	▲ E	L	a	b	▲ E
0	-64.47 ± 2.75	11.17 ± 1.98	11.87 ± 1.81		-64.09 ± 1.86	10.92 ± 1.95	11.19 ± 1.56	
1	-61.62 ± 1.58	11.50 ± 2.13	11.96 ± 1.72	2.87	-61.22 ± 1.91	11.17 ± 2.22	10.49 ± 1.72	2.97
2	-62.75 ± 1.81	10.18 ± 2.03	11.03 ± 1.66	2.15	-62.58 ± 1.71	9.36 ± 1.84	9.33 ± 1.94	2.86
3	-62.92 ± 2.09	11.02 ± 2.53	11.76 ± 2.02	1.56	-63.95 ± 1.87	10.19 ± 1.74	10.74 ± 1.77	0.87
4	-62.44 ± 1.54	11.31 ± 2.30	11.90 ± 1.63	2.03	-62.90 ± 1.16	10.82 ± 1.87	10.86 ± 1.71	1.24
7	-63.16 ± 1.18	10.52 ± 2.15	11.78 ± 1.80	1.46	-63.66 ± 1.04	11.12 ± 1.92	11.76 ± 1.66	0.74
10	-60.17 ± 0.93	8.95 ± 1.74	8.82 ± 1.04	5.72	-60.34 ± 0.86	9.00 ± 1.53	8.06 ± 1.34	5.25
14	-62.88 ± 1.14	10.56 ± 1.81	12.01 ± 1.73	1.70	-62.94 ± 1.24	10.21 ± 1.65	11.35 ± 1.61	1.35

Table 3

Effects of HVEF on the Hunters color value of tomato fruit during 1 week post RF treatment shelf life test

treatment interval (days)	HVEF sample				Control sample			
	L	a	b	▲ E	L	a	b	▲ E
1	-62.12 ± 1.06	10.31 ± 1.64	10.68 ± 1.61	1.55	-63.03 ± 1.13	10.54 ± 1.68	11.29 ± 1.43	0.34
2	-62.53 ± 1.16	10.73 ± 2.00	12.20 ± 1.67	0.43	-62.22 ± 1.58	11.52 ± 1.81	12.65 ± 1.79	1.98
3	-61.82 ± 1.27	11.10 ± 1.93	12.63 ± 1.72	1.34	-62.28 ± 1.09	10.89 ± 1.69	11.72 ± 1.46	1.02
4	-60.06 ± 1.04	9.11 ± 1.74	10.03 ± 1.44	3.74	-59.68 ± 0.82	9.62 ± 1.77	9.85 ± 1.58	3.65
7	-59.21 ± 1.04	9.02 ± 1.54	9.68 ± 1.63	4.61	-59.42 ± 0.70	9.26 ± 1.29	9.48 ± 1.16	4.10

3.1.3 Effect of HVEF on Total Soluble Sugar, TSS (°Brix)

High °Brix concentration were observed for continuous electric field sample treatment for most days during the 2 weeks of treatment when compared with control (as shown in Figure 4). The TSS content of electric field samples were in range of 6.48 to 6.78 °Brix while control was in range of 6.41 to 6.87 °Brix. However on day 1 and 3 the control samples had slightly higher °Brix content of 6.79 and 6.87 °Brix. For post electric field shelf life testing period the electric field treated sample °Brix was overall higher with highest value recorded on day 7 of 7.06 compared to control. As shown in Figure 5 a continuous increase in °Brix content trend was seen with electric field sample whilst control sample °Brix showed a fluctuating trend.
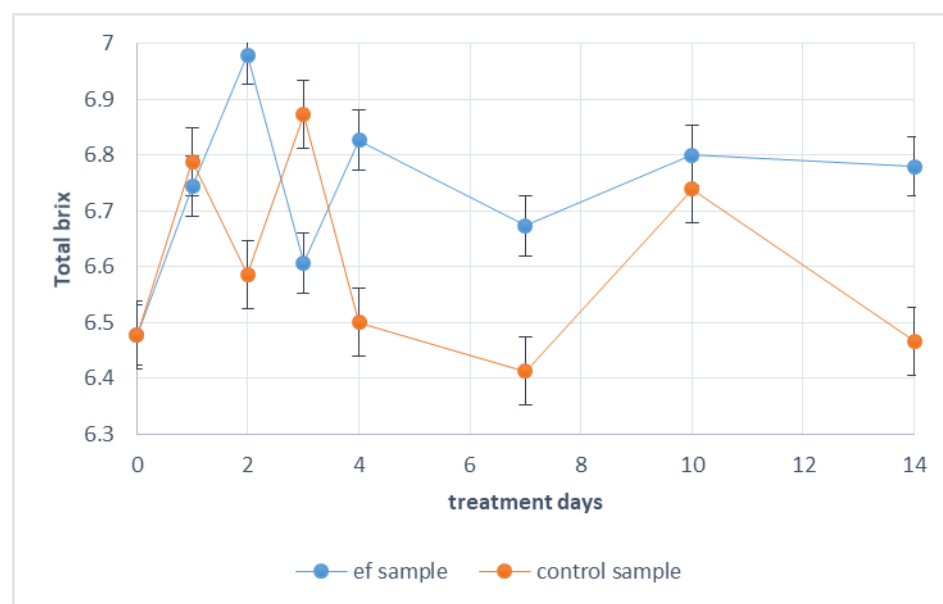

Fig. 4 Measured Brix values of continuous HVEF and control samples of 14 days of storage period (1⁰C; 95% RH)
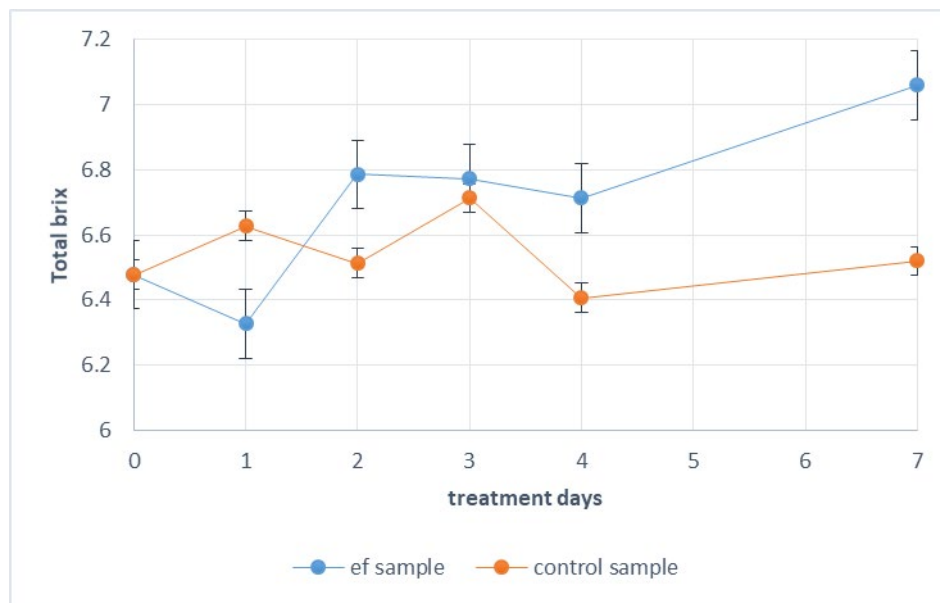
Fig. 5 Measured Brix values of post HVEF treatment and control samples of 7 days of shelf life storage testing period (1^0C; 95% RH)

3.1.4 Effect of HVEF on fruit firmness

Evaluating firmness is one of the important characteristic to determine the fruit texture and degree of ripeness or maturity of fruits and vegetables. Results from continuous electric field treatment as shown by Figure 6 indicated that electric field treatment did not have a significant effect on the fruit firmness. Overall the control sample tend to had more firm fruit texture except on day 4 and 7 where the EF treated samples were slightly firmer. The highest EF sample was 5.37N on day 4 while for control (5.87N) was the highest on day 2. For post electric field shelf life testing period the EF treated sample showed more firmness overall however the difference in firmness level was only slight compared to the control.
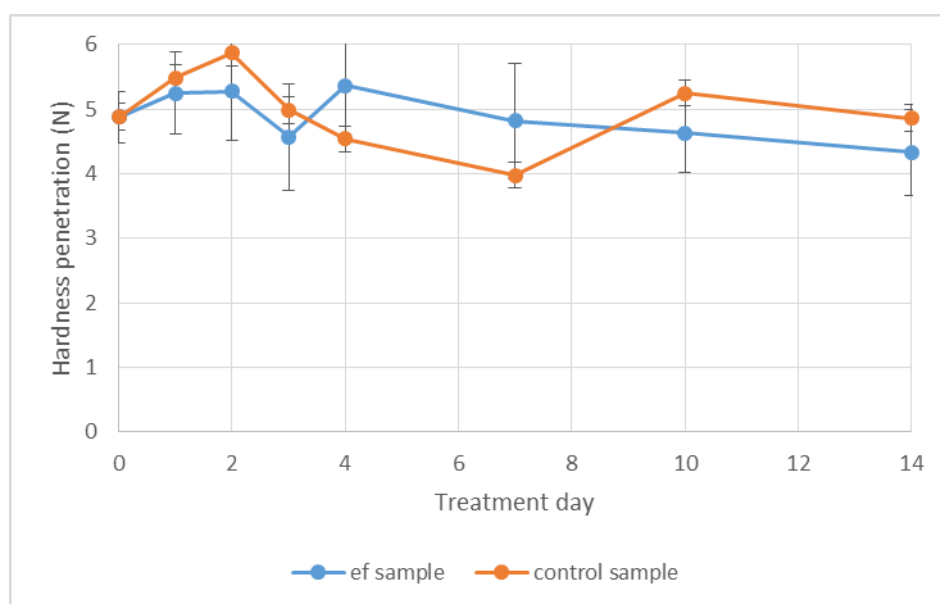
Fig.6 Fruit firmness level (N) of continuous HVEF and control samples of 14 days of storage.

3.1.5 Effect of HVEF on fruit respiration (CO_2 yield).

According to (Chi-En Liu., 2017) the rate at which the carbon dioxide gas is produced from the fruit can be used as an important physical parameter to determine the ripeness level of a fruit. In addition as described by (Bhande, 2008) that the rate at which the CO_2 gas produced has a direct impact on the life span of the fruit after harvesting. Its also described that CO_2 level is a good indicator of shelf life as the higher the rate of CO_2 production; the shorter is the shelf life. In this experiment the CO_2 production was measured by allowing CO_2 gas to accumulate inside the air tight dessicator. The CO_2 level was measured every morning corresponding to the planned monitoring schedule during the 2 weeks of continuous treatment.

As shown by Figure 7, samples stored under 1^oC had the highest production of CO_2 gas recorded followed by samples stored under 0^oC for both the EF treated sample and the control. Samples that were stored under 3^oC was found to be in medium range of CO_2 production. The least CO_2 yield was seen for sample stored under 5^oC. The samples stored under 5^oC had significantly lower CO_2 production overall. Observations under this treatment temperature, shows that samples treated to continuous electric field treatment had slightly high CO_2 production compared to control.
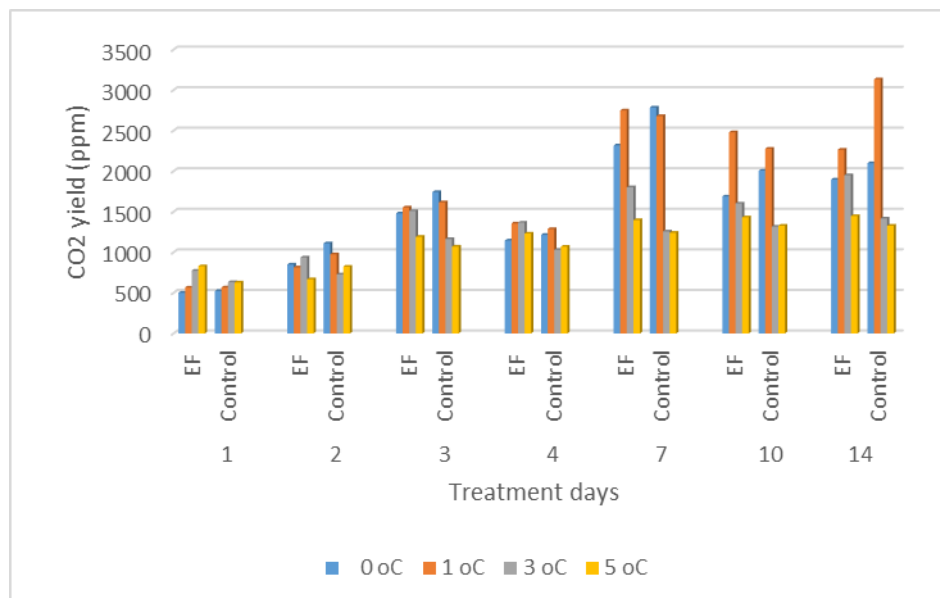

Fig 7 Carbon dioxide yield in ppm during 2 weeks of continuous electric field treatment

Post electric field treatment samples (Figure 8) that were stored under 3^oC showed least percent of CO_2 production. It was also very clearly evident that samples exposed to continuous electric field treatment had suppressed CO_2 yield on all of the days. Similar trends were also observed with samples stored at 0^oC and 1^oC respectively.
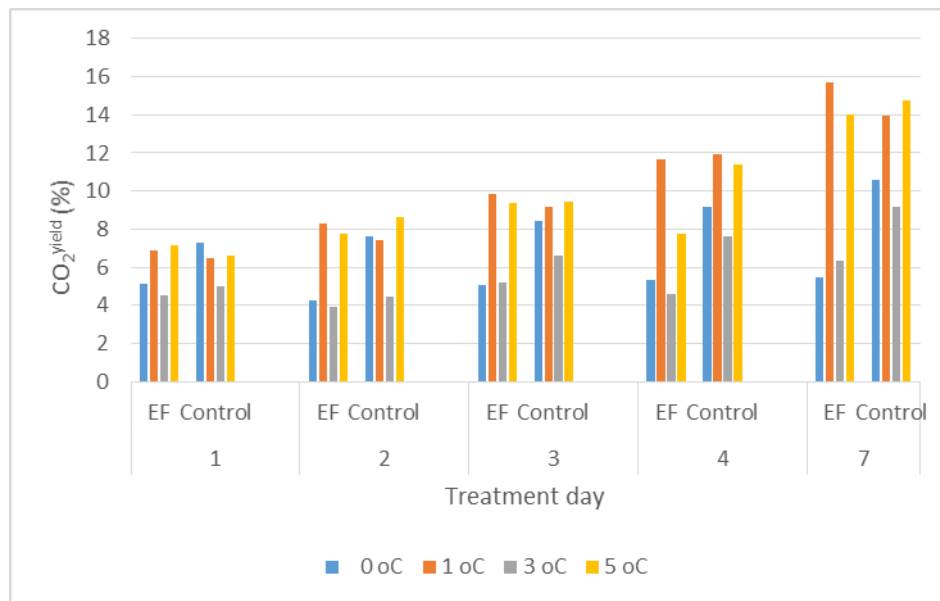
Fig 8 Carbon dioxide yield in % during 1 weeks of post electric field shelf life testing period

4. CONCLUSION

In conclusion 2 distinct type of electric field set up was used to study quality attributes and respiration rate on the shelf life extension of tomato fruits. The quality attributes were measured using a 7kV large incubator while the respiration was measured using 2kV aluminum parallel plate setup. The results showed that both type of electric field set up has a positive effect on extending the shelf life of tomato fruits. The 7kV electric field treated samples showed suppressed weight loss, firmer texture and higher Brix level during shelf life storage period. For Respiration results it was seen that at 3°C the CO_2 was least during 1 week shelf life testing of electric field treated samples. It can be said that HVEF is effective in maintaining tomato fruits quality and freshness.

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11:00 AM - 11:15 AM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Hall A)

[4-1015-A-04] Impact of Low Electric Field on Physicochemical Properties and Antioxidant Activity of Persimmon (*Diospyros kaki*)

Naruesorn Jaisue[1,2], Sutthiwal Setha[1,2], Daisuke Hamanaka[3], *Matchima Naradisorn[1,2] (1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 2. Research Group of Postharvest Technology, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 3. Faculty of Agriculture, Kagoshima University, Kagoshima, Japan(Japan))
Keywords: Electric field, Non-thermal technology, Persimmon, Total phenolic content, Antioxidant activity

Electric field technology is a mild preservation technology that has been recently applied to maintain nutritional and physicochemical properties of food products; and improve juice yield and polyphenol extraction in fruits. In fresh fruit, high electric field treatment showed its efficacy in suppressing respiration rate of pear, plum and banana; and delaying ripening of mature green banana and sweet pepper. However, electric field with high voltage affects texture of some fruits and vegetables by causing loss of turgor and crack of cell membranes. The use of low electric field (LEF) may be of interest as an effective tool to maintain postharvest quality of fresh fruits without causing defects. The aim of this study was to investigate the effect of low electric field (LEF) on physicochemical properties and antioxidant activity of persimmon (*Diospyros kaki*). Persimmon fruit were exposed to electric field strength of 7 kV/cm during storage at 10 ˚ C. Persimmons stored at 10 ˚ C without LEF treatment were used as a control. Analyses were performed at 3-day intervals during storage. The results showed that the firmness of persimmon fruit treated with LEF during storage for 9 days were significantly higher ($P <0.05$) than those in the non-LEF storage condition. Exposure of LEF for 3 and 9 days significantly increased ($P <0.05$) total phenolic content and antioxidant activity in persimmons. There was no significant difference in weight loss, colour (L^*, a^*, b^* values) and respiration rate among LEF-treated and untreated persimmons. After 15 days of storage, application of the LEF treatment for 9 days prior to storage at 10 ℃ significantly increased ($P <0.05$) total phenolic content in pulp and peel by 55.76% and 41.09%, respectively, in comparison to the non-LEF treatment. This study suggests that exposure to low electric field strength during storage may be useful for prolonging shelf life of persimmon; and for producing and preserving persimmon product with high total phenolic content and antioxidant capacity.

11:15 AM - 11:30 AM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Hall A)

[4-1015-A-05] Effects of Ultrasound Pretreatments on the Texture and Colour Kinetics of Sweet Potato (Ipomea batatas) during Deep Fat Frying

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Keywords: frying, ultrasound, kinetics, texture, colour, sweet potato

The effects of ultrasound pretreatment prior to frying on the texture and colour kinetics of deep fat fried sweet potato were investigated. Three different pretreatment methods (UD-ultrasound with distilled water, OD-Osmotic dehydration without ultrasound and UOD-ultrasound assisted osmotic dehydration) and the untreated (control) were employed in the experiment. Pretreated and untreated samples were fried in a deep

fat fryer at 130-170 $^{\circ}$C for 2-10 min. The kinetic models for texture and colour fitted the experimental data with high coefficient of determination (> 0.700). The normalized maximum force (F*) of samples pretreated by UD showed no significant difference with that of the untreated, while samples pretreated by UOD and OD were significantly higher than that of the untreated. The rate of hardening process was significantly enhanced in sweet potato pretreated by UOD and OD during frying. For colour, samples pretreated by UOD showed high lightness (L/L$_o$), low redness (a/a$_o$), moderate yellowness (b/b$_o$) and low total colour difference (Δ E/ Δ E$_o$) compared to other pretreatment methods at 150 $^{\circ}$C. The rate of formation of lightness and yellowness of fried sweet potato was enhanced in samples pretreated by UOD. The texture and colour kinetics of pretreated and untreated fried samples were temperature dependent and expressed as activation energy. Therefore, ultrasound pretreatment is a good technology that could be applied to positively influence the texture and colour of sweet potato during deep fat frying.

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1015-A] Postharvest/Food Technology and Process Engineering (1)

Wed. Sep 4, 2019 10:15 AM - 12:00 PM Hall A (Main Hall)

[4-1015-A-06] Effect of Electromagnetic Field Pretreatment on Selected Vitamins and Fibre of Sweet Pepper and Fluted Pumpkin Leaf

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Keywords: Fluted Pumpkin Leaf, Magnetic Field, Nutrients, Electromagnetism, Sweet Pepper, Pretreatment, Blanching

Sweet pepper (SP) and Fluted pumpkin leaf (FPL) are vegetables that contain vitamins and fibre. SP has contents that cure lung cancer, diabetes, cataracts, rheumatism, arthritis, fever, cold, sores and bruises. FPL can improve hematological parameters, has anti-inflammatory and anticholesterolemic properties. Thermal pretreatment (heat addition) of vegetables during processing has the tendency of reducing their qualities. One of the promising non-thermal methods of pretreatment that is novel/emerging/non-conventional is the use of Magnetic Field (MF); however, the method is still grossly underutilized. Therefore, the effect of electromagnetic field types (static, pulse and alternating), magnetic field strength (5–30 mT) and pretreatment time (5-25 min) on the vitamins (C and A) and fibre of SP and FPL were investigated. An existing, but new electromagnetic field pretreatment device was used to conduct the experiment designed with Design Expert software (6.0.6 version). All electromagnetic field pretreated samples, controls (blanched and fresh) were analyzed in for vitamin C, vitamin A and fibre. Results showed that most of the electromagnetic field pretreatment retained/improved the vitamins (C and A) and fibre better than blanching. Vitamin C of SP and FPL are 73 - 85 mg/100g and 60 - 61.50 mg/100g for the electromagnetic field pretreatment and the blanched was 71 and 60 mg/100g for SP and FPL respectively. Vitamin A of SP and FPL were 0.0058 - 0.008 mg/100g and 0.002 mg/100g – 0.003 mg/100g for the electromagnetic field pretreatment, but blanched samples had about 0.001 mg/100g. Electromagnetic field pretreatment caused the fibre of SP and FPL to be between 1.7- 2.6% and 6 - 10% and blanched to be 1.8 and about 7% respectively. Results indicated `that electromagnetic field pretreatment of vegetables is a possible replacement alternative for blanching.

Effect of Electromagnetic Field Pretreatment on Selected Vitamins and Fibre of Sweet Pepper and Fluted Pumpkin Leaf

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ABSTRACT

Sweet pepper (SP) and Fluted pumpkin leaf (FPL) are vegetables that contain vitamins and fibre. SP has contents that cure lung cancer, diabetes, cataracts, rheumatism, arthritis, fever, cold, sores and bruises. FPL can improve hematological parameters, has anti-inflammatory and anticholesterolemic properties. Thermal pretreatment (heat addition) of vegetables during processing has the tendency of reducing their qualities. One of the promising non-thermal methods of pretreatment that is novel/emerging/non-conventional is the use of Magnetic Field (MF); however, the method is still grossly underutilized. Therefore, the effect of electromagnetic field types (static, pulse and alternating), magnetic field strength (5–30 mT) and pretreatment time (5-25 min) on the vitamins (C and A) and fibre of SP and FPL were investigated. An existing, but new electromagnetic field pretreatment device was used to conduct the experiment designed with Design Expert software (6.0.6 version). All electromagnetic field pretreated samples, controls (blanched and fresh) were analyzed in for vitamin C, vitamin A and fibre. Results showed that most of the electromagnetic field pretreatment retained/improved the vitamins (C and A) and fibre better than blanching. Vitamin C of SP and FPL are 73 - 85 mg/100g and 60 - 61.50 mg/100g for the electromagnetic field pretreatment and the blanched was 71 and 60 mg/100g for SP and FPL respectively. Vitamin A of SP and FPL were 0.0058 - 0.008 mg/100g and 0.002 mg/100g – 0.003 mg/100g for the electromagnetic field pretreatment, but blanched samples had about 0.001 mg/100g. Electromagnetic field pretreatment caused the fibre of SP and FPL to be between 1.7- 2.6% and 6 - 10% and blanched to be 1.8 and about 7% respectively. Results indicated `that electromagnetic field pretreatment of vegetables is a possible replacement alternative for blanching.

Keywords: Fluted Pumpkin Leaf, Magnetic Field, Nutrients, Electromagnetism, Sweet pepper, Pretreatment, Blanching

1. INTRODUCTION

Sweet pepper (SP) and fluted pumpkin leaf (FPL) are vegetables scientifically referred to as *Capsicum annum* and *Telfairia occidentalis* respectively. They both contain vitamins, fibres and other nutrients needed for growth, development and maintenance of human body. Some reported health benefits of sweet pepper are its ability to cure the following ailments: lung cancer, arthritis, diabetes, cataracts, rheumatism, fever and cold (Odewole and Olaniyan, 2015). Fluted pumpkin leaf can improve hematological parameters, has anti-inflammatory and anticholesterolemic properties (Obeagu et al., 2014).

Pretreatment is one of the unit operations in vegetable processing value chain. It is done primarily to aid the overall qualities of vegetables in terms of improvement/retention of nutritional, sensory and functional properties as well as reduction of microbial load. It is a modification unit operation and the extent of modification is governed by factors such as: method of pretreatment, duration of pretreatment and other properties(intrinsic/extrinsic) of the food to be pretreated. Neeto and Chen (2014) classified food pretreatment/processing methods into conventional and non-conventional. One

of the major characteristics of the conventional method is its dependency on high temperatures to achieve the objective of the pretreatment.

The non-conventional method of pretreatment which is sometimes referred to as novel or emerging pretreatment technology does not necessarily need high temperature. Some typical examples of non-conventional pretreatment method that are non-thermal are: Pulsed Electric Field (PEF), High Hydrostatic Pressure (HPP), irradiation, pulsed light (Neeto and Chen, 2014) and magnetic field. Non-conventional pretreatment method can also take place under sub-lethal or room temperature (Pereira and Vincente, 2010) that would not cause significant negative effects on product qualities. Also, assurance of food of better quality is high with non-thermal pretreatment (Barbosa-Canovas et al., 2005).

Blanching is a widely used method of pretreating fruits and vegetables. It is one of the conventional thermal (heat addition) processing methods of food. It involves dipping food materials in hot water (at 100 °C or slightly below) or other food grade liquids; holding it for a short time (few seconds to few minutes depending on the type and condition of the food); removal of the food from hot medium; and rapidly quenching the hot food with cold water to avoid cooking. Blanching can have negative effect on food nutrients such, as vitamins and phenolic compounds which are relatively unstable when subjected to heat treatments (Prochaska et al., 2000). The method has led to the degradation and leaching of nutritive components, such as sugars, minerals and vitamins of food during processing (Cumming et al., 1981; Rincon et al., 1993; Vidal-Valverde and Valverde 1993). Nobosse et al. (2017) reported that steam blanching caused reduction in vitamin C of moringa leaf. Higher blanching time and higher temperature negatively affected the vitamin C of three blanched leafy vegetables (Olayinka et al., 2012). Vitamin C (ascorbic acid) retention level was said to be the index for judging the overall nutrient retention ability of blanched food products (Murcia et al., 2000). The reason for this is that ascorbic acid (vitamin C) is the least stable nutrient during processing; it is highly sensitive to oxidation and leaching into water-soluble media during processing of food (Franke et al., 2004 in Patras et al., 2011).

Barbosa-Canovas et al. (2005) defined magnetic field as a region of space in which a magnetic body is capable of inducing surrounding bodies. Electromagnetism is the generation of magnetic field due to the flow of current in a conductor. In 1985, the use of magnetic field as a non-thermal method of preserving food was proposed for the first time (Barbosa-Canovas et al., 2005). When harvested vegetables are placed within magnetic field, the external magnetic field from either permanent or temporary magnetic (electromagnet) will interact with their living cells. This interaction will lead to modifications responsible for quality improvement. The guidelines given on the acceptable limit of exposure of different parts of human beings to magnetic field are 2-8T for head, trunk and limbs, and 400 mT for any part of the body (ICNIRP, 2009).

Magnetic field for food processing and related areas exist (Hayder et al., 2015, Ibara et al., 2015, Lipiec et al., 2004, Ordonez and Berrio 2011, Jia et al. 2015 and Kyle, 2015). However, available information is very few and sketchy on the use of magnetic field for vegetable processing and on nutritional qualities of pretreated foods. This is a clear indication that magnetic field is still grossly underutilized and its excellent advantages are yet to be substantially harnessed in food processing. Therefore, the objective of this study was to investigate the effect of three types of magnetic fields (generated by a developed electromagnetic field (EF) pretreatment device); magnetic field strength and pretreatment time on vitamin C, vitamin A and fibre of sweet peeper (SP) and fluted pumpkin leaf (FPL).

2. MATERIALS AND METHODS
2.1 Materials
An electromagnetic field pretreatment device; electronic weighing balance (OHAUS, Model 201, China), fresh samples of sweet pepper and fluted pumpkin leaf.
2.2 Experimental Procedures
Fresh samples of sweet pepper (SP) and fluted pumpkin leaf (FPL) of good quality were procured, sorted, washed and cut into pieces in order to ensure better exposure to the magnetic field pretreatment. After these, uniform experimental quantities per run (10 g for FPL and 100 g for SP) were measured with an electronic weighing balance (OHAUS, Model 201, China). The measured samples were placed in the electromagnetic field device. Selection of magnetic field types (Static, Pulse or Alternating)

with combination of magnetic field strength (5-30 mT) and pretreatment time (5-30 min) (set with timer on the device) was executed according to the experimental design and layout done with Design Expert software (version 6.0.6) as shown in Table 1.

Blanched and fresh samples were used as controls. After pretreatment, all samples were taken to the laboratory for analysis. AOAC (2005) standard procedure was used to analyze vitamin C, vitamin A and fibre content of all the samples.

Table 1. Experimental design and layout

SN	SMF/PMF/AMF				
	SP		FPL		
	MFS (mT)	PT (min)	MFS (mT)	PT (min)	Outputs
1	13.5 **(9.5)**	15 **(5)**	13.5 **(9.5)**	15 **(5)**	
2	19.0 **(5.0)**	20 **(25)**	19.0 **(5.0)**	20 **(25)**	
3	19.0 **(9.5)**	15 **(25)**	19.0 **(9.5)**	15 **(25)**	
4	19.0 **(9.5)**	15 **(15)**	19.0 **(9.5)**	15 **(15)**	
5	19.0 **(5.0)**	15 **(15)**	19.0 **(5.0)**	15 **(15)**	
6	8.0 **(9.5)**	25 **(15)**	8.0 **(9.5)**	25 **(15)**	
7	8.0 **(14.0)**	5 **(5)**	8.0 **(14.0)**	5 **(5)**	
8	19.0 **(9.5)**	15 **(15)**	19.0 **(9.5)**	15 **(15)**	
9	19.0 **(9.5)**	10 **(15)**	19.0 **(9.5)**	10 **(15)**	
10	24.5 **(14.0)**	15 **(15)**	24.5 **(14.0)**	15 **(15)**	
11	30.0 **(5.0)**	25 **(5)**	30.0 **(5.0)**	25 **(5)**	
12	30.0 **(9.5)**	5 **(15)**	30.0 **(9.5)**	5 **(15)**	
13	19.0 **(14.0)**	15 **(25)**	19.0 **(14.0)**	15 **(25)**	

SMF-Static Magnetic Field; **PMF**- Pulse Magnetic Field; **AMF**-Alternating Magnetic Field; **Outputs** are vitamin C, vitamin A and fibre content.
Note: combination of values in brackets were used for AMF experiment and those not in brackets were used for SMF and PMF experiments.

3. RESULTS AND DISCUSSION
3.1 Effect of Electromagnetic Field Pretreatment on the Vitamins C and A of SP and FPL

The illustrations of the effect of the electromagnetic field pretreatment on the vitamin C of SP and FPL are presented in Figures 1–4. For SP, all the electromagnetic field (EF) pretreatment combinations retained vitamin C (80 mg/100g) than blanched sample (about 70 mg/100g) with fresh sample having 74 mg/100g. Similarly, for FPL, almost all the EF pretreatments have vitamin C values higher than blanched (slightly above 60 mg/100g) and fresh samples (slightly above 60 mg/100g). The highest values of vitamin C (61.5 mg/100g) was obtained at PMF-7(24.5 mT and 15 min). Improvement/better retention of vitamin C under electromagnetic field pretreatment could be due to the absence or very mild presence of heat (Pereira and Vincente, 2010) during pretreatment. However, water blanching which is always associated with high pretreatment temperature (not less than 90^0C) caused vitamin C which is heat sensitive, water soluble and least stable during processing (Patras *et al.*,2011) to be depleted. Vitamin C of about 80 mg/100g and 120 mg/100g were obtained after pretreating onion and red bell pepper (sweet pepper) with osmotic solution of salt and drying at 60^0C (Alabi et al., 2016, and Odewole and Olaniyan, 2016). Blanching pretreatment also degraded the ascorbic acid (vitamin C) of *colocasia* vegetable by 43.12% - 63.19% (Kaushal et al.,2013).

Furthermore, almost all the electromagnetic field pretreatment caused the vitamin A content of SP to have between 0.0055 mg/100g to 0.008 mg/100g, but blanched and fresh samples have about 0.0055 mg/100 and 0.0078 mg/100g respectively. Similarly, for FPL, the vitamin A content was about 0.002 mg/100g – 0.003 mg/100g for most of the electromagnetic field pretreatment; fresh and blanched samples have about 0.002 and 0.001 mg/100g vitamin A respectively. These observations showed that electromagnetic field pretreatment (a non-thermal method) caused retention/improvement of vitamin A better than blanching pretreatment (a thermal method). Odewole and olaniyan (2016) obtained about

1.40 mg/100g vitamin A for red bell pepper (same as sweet pepper) pretreated with osmotic solution of salt and dried at 60^0C. Vitamin A content of mango chips pretreated with osmotic solution of sugar was found to be about 6.46 mg/100g (Odewole et al., 2014).
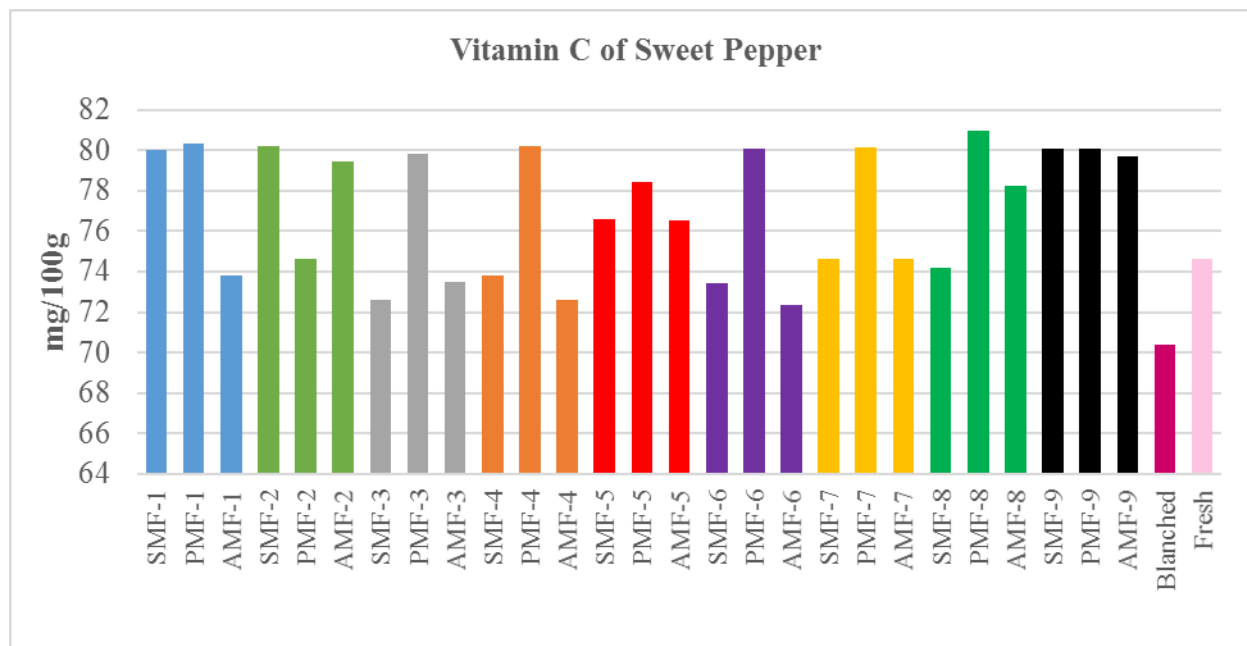

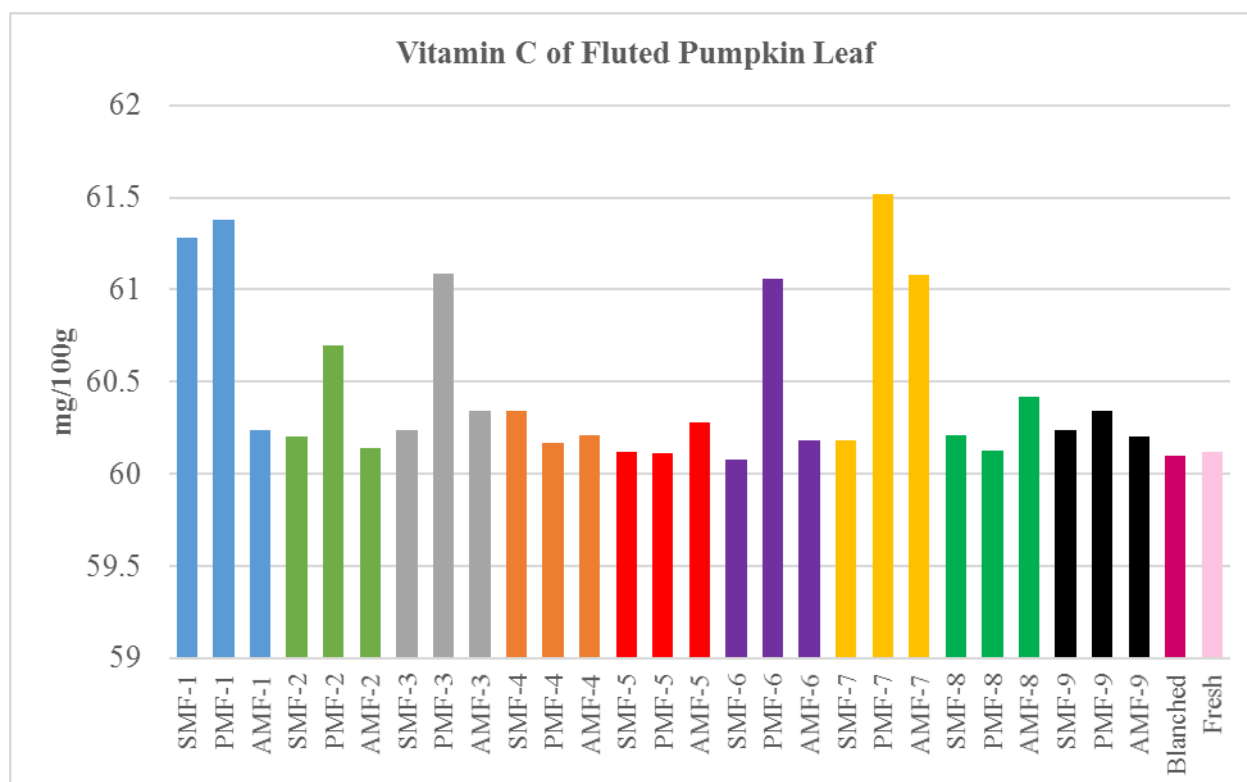Figure 1. Effect of EF pretreatment on Vitamin C of sweet pepper

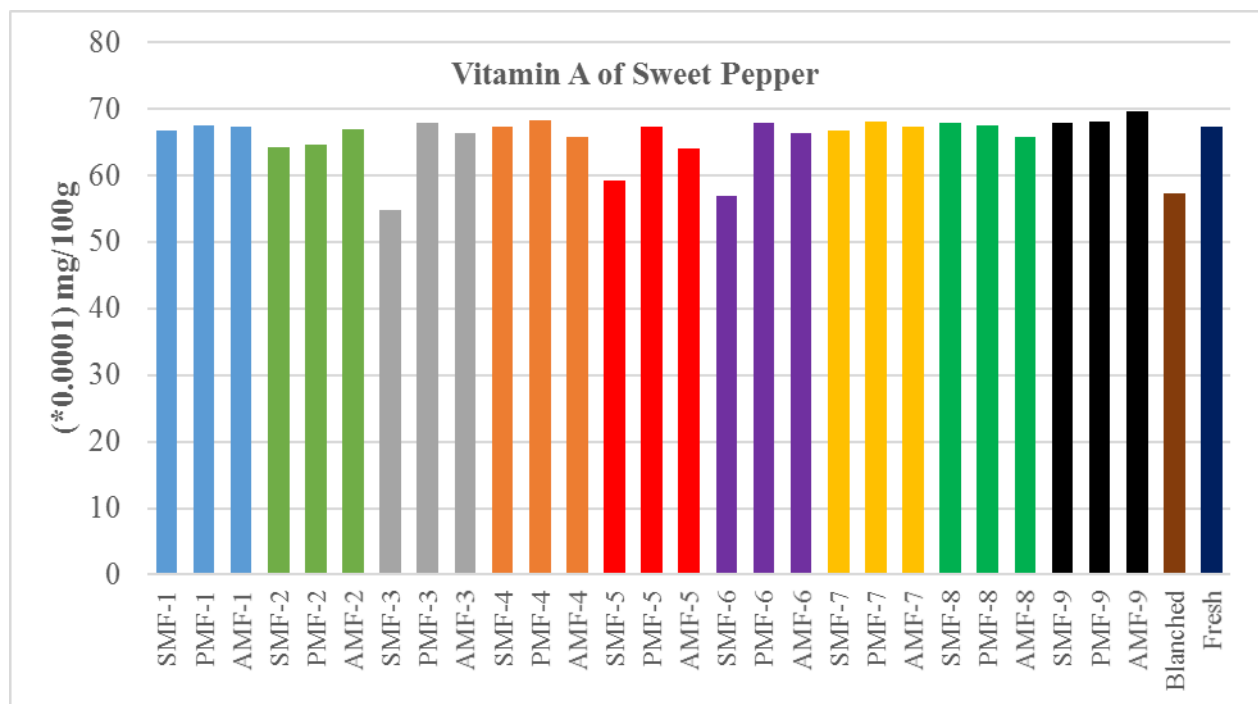Figure 2. Effect of EF pretreatment on Vitamin C of fluted pumpkin leaf

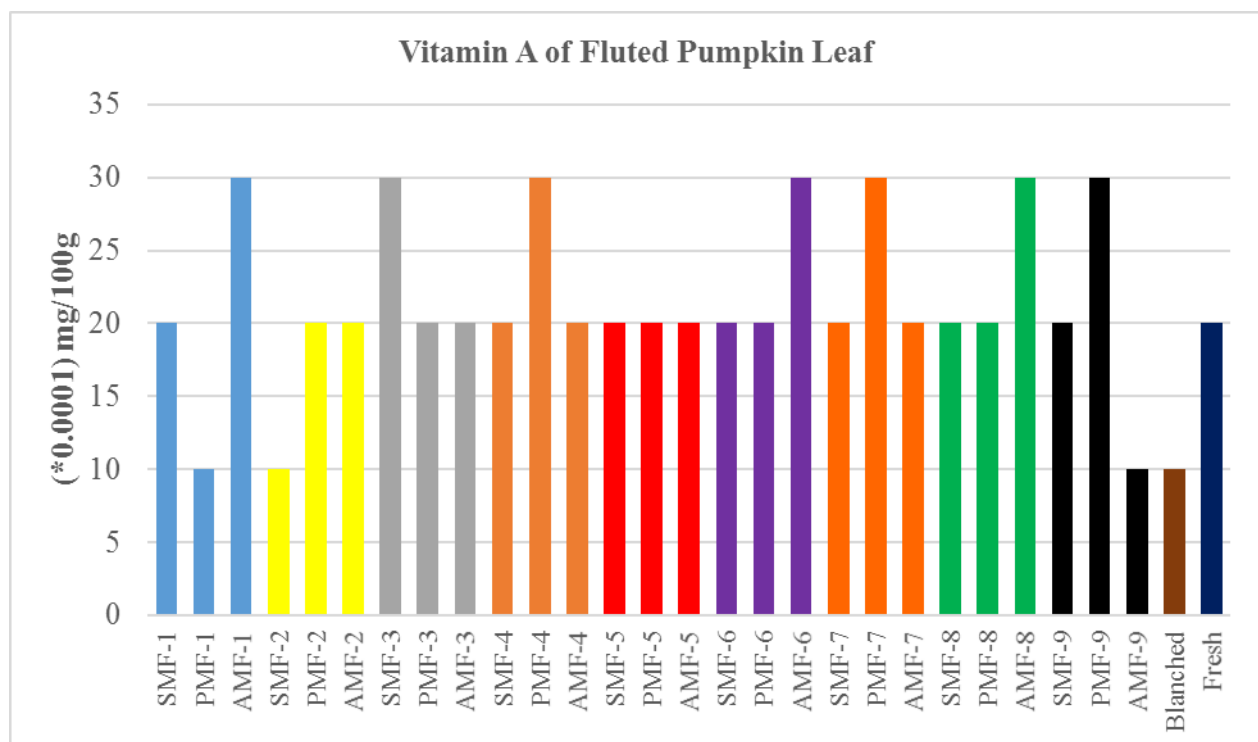Figure 3. Effect of EF pretreatment on Vitamin A of sweet pepper

Figure 4. Effect of EF pretreatment on Vitamin A of fluted pumpkin leaf

3.2 Effect of Electromagnetic Field Pretreatment on the Fibre of SP and FPL

Figures 5 and 6 show the effect of electromagnetic field (EF) pretreatment on the fibre content of SP and FPL. Many EF pretreated and fresh samples of SP have fibre content more than the blanched sample. Highest value of about 2.6% of fibre was obtained for SP at SMF-3 (19 mT and 15 min), AMF-7(14 mT and 15 min) and SMF-8(19 mT and 15 min). For FPL, almost all the EF pretreated samples have fibre content higher than blanched and fresh samples. PMF-1 (13.5 mT and 15 min),

AMF-8 (5 mT and 5 min) and PMF-9 (30 mT and 5 min) have highest fibre value of 10% while blanched and fresh samples have about 7.5% and 7% respectively. The fibre content of sweet pepper was about 8% after pretreating with osmotic solution of salt and drying at 60^0C (Odewole and Olaniyan, 2015).
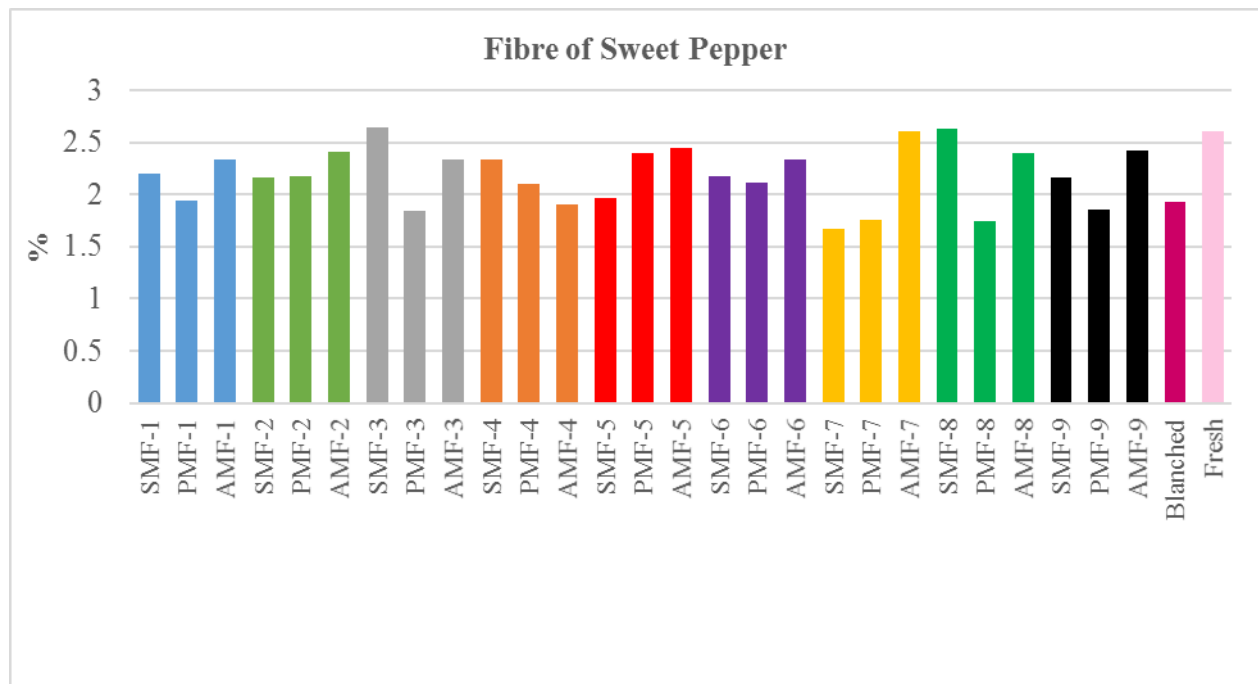
Figure 5. Effect of EF pretreatment on Fibre of sweet pepper
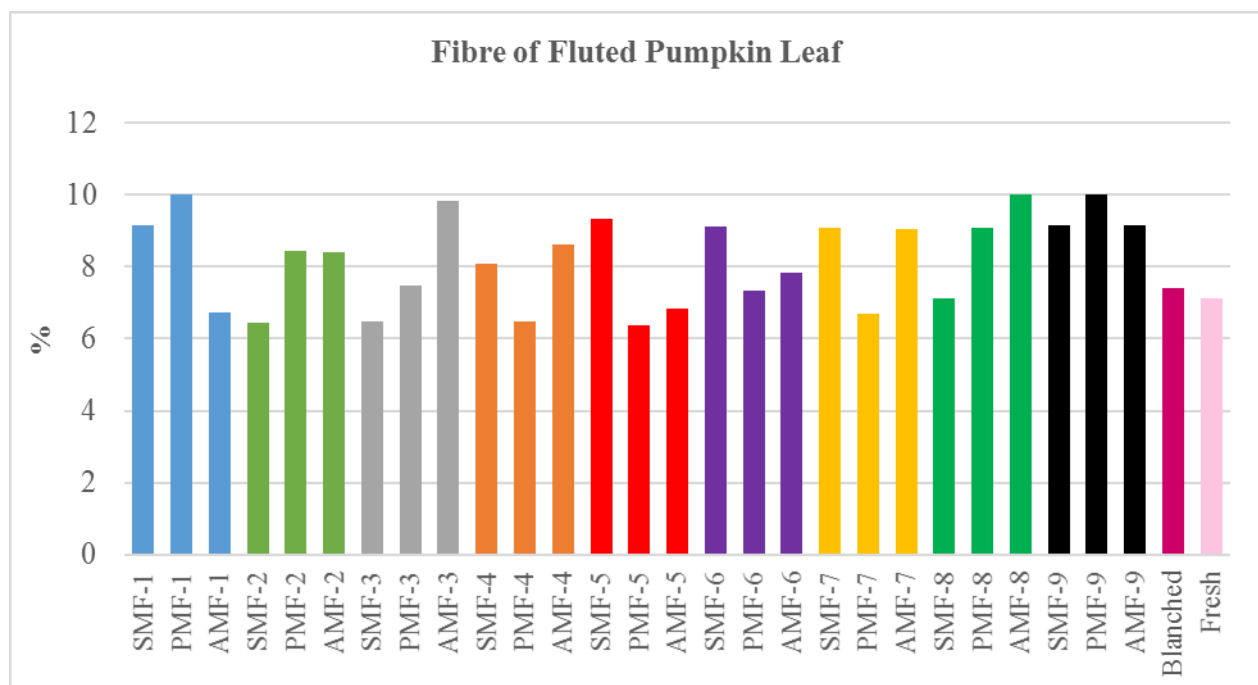
Figure 6. Effect of EF pretreatment on fibre of fluted pumpkin leaf

4. CONCLUSION AND RECOMMENDATION
Electromagnetic field pretreatment in most cases led to the improvement/better retention of vitamin C, vitamin A and fibre of sweet pepper and fluted pumpkin leaf than blanching. Therefore,

electromagnetic field pretreatment (a non-thermal method) is a promising pretreatment alternative that can be explored for replacing blanching (thermal method) in vegetable processing value chain. Modelling and optimization of the process is highly recommended for future research.

ACKNOWLEDGMENT

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Oral Session | Postharvest/Food Technology and Process Engineering

[4-1015-A] Postharvest/Food Technology and Process Engineering (1)

Wed. Sep 4, 2019 10:15 AM - 12:00 PM Hall A (Main Hall)

[4-1015-A-07] Pulsed electric fields applications in fresh meat

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Keywords: meat, pulsed electric field, non-thermal, tenderness, quality

Pulsed electric fields (PEF) is a novel minimal processing technology that is applied to produce high quality food products with a natural flavour and fresh appearance and is gaining recognition due to the increasing demands of consumers for fresh and natural foods. The technology involves the application of an external electric field to induce electroporation, and formation of pores that increases the membrane and cellular permeability. Over the last 6 years, there has been much interest to use PEF to modify the properties of fresh meat. Our lab was the first to demonstrate the potential use of PEF in meat tenderization and achieved about 20% increase in tenderness of longissimus and semimembranosus beef muscles using various PEF processing intensities. This improvement in tenderness did not affect other major quality attributes such as colour and lipid oxidation. Further research from our lab demonstrated an early activation of Calpains, calcium activated proteases that are naturally found in the muscles and are involved in postmortem degradation of structural proteins. Recently, we used PEF to accelerate the dry aging of meat and demonstrated the ability to reduce the processing time to about half of its normal value. Further, the technology was also found to influence the diffusion, distribution and release of sodium from meat matrix and thus could be utilized as tool to reduce the sodium in processed meat products. This presentation will highlight the various potential applications of PEF in fresh meat and discuss the mechanisms responsible for the potential positive outcomes in PEF treated meat. Furthermore, negative aspects of PEF use will be also discussed with the aim of providing complete information on PEF application in fresh meat processing for proper evaluation of commercial use of the technology.

Pulsed electric field applications in fresh meat

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ABSTRACT

Pulsed electric fields (PEF) is a novel minimal processing technology that is applied to produce high quality food products with a natural flavour and fresh appearance and is gaining recognition due to the increasing demands of consumers for fresh and natural foods. The technology involves the application of an external electric field to induce electroporation, and formation of pores that increases the membrane and cellular permeability. Over the last 6 years, there has been much interest to use PEF to modify the properties of fresh meat. Our lab was the first to demonstrate the potential use of PEF in meat tenderization and achieved about 20% increase in tenderness of longissimus and semimembranosus beef muscles using various PEF processing intensities. This improvement in tenderness did not affect other major quality attributes such as colour and lipid oxidation. Further research from our lab demonstrated an early activation of Calpains, calcium activated proteases that are naturally found in the muscles and are involved in postmortem degradation of structural proteins. Recently, we used PEF to accelerate the dry aging of meat and demonstrated the ability to reduce the processing time to about half of its normal value. Further, the technology was also found to influence the diffusion, distribution and release of sodium from meat matrix and thus could be utilized as tool to reduce the sodium in processed meat products. This presentation will highlight the various potential applications of PEF in fresh meat and discuss the mechanisms responsible for the potential positive outcomes in PEF treated meat. Furthermore, negative aspects of PEF use will be also discussed with the aim of providing complete information on PEF application in fresh meat processing for proper evaluation of commercial use of the technology.

Key words: meat, pulsed electric field, non-thermal, tenderness, quality.

Oral Session | Food Safety

[4-1015-C] Food Safety (1)

Chair:Anthony Mutukumira(Massey University, New Zealand), siti nurjanah(Bogor Agricultural University)
Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room C (3rd room)

[4-1015-C-01] **Surface Pasteurisation of Fresh Chicken Meat using UV-C Technology**

Arthur Jonathan Philip[1], Negah Nikanjam[1], Emilia Nowak[1], *Anthony Mutukumira[1]　(1. Massey University(New Zealand))

10:15 AM - 10:30 AM

[4-1015-C-02] **Efficient Filtering of Live *Escherichia coli* by Using 60 GHz CMOS Sensor**

*Hiroki Fukuda[1], Tetsuhito Suzuki[1], Naoshi Kondo[1], Yuichi Ogawa[1]　(1. Graduate School of Agriculture, Kyoto University(Japan))

10:30 AM - 10:45 AM

[4-1015-C-03] **Safety Evaluation of Bacteriocinogenic Strains of *Pediococcus acidilactici* Isolated From Artisanal Cheeses**

Luis Augusto Nero[1], Yosep Ji[2], Wilhelm Holzapfel[2], *Svetoslav Dimitrov Todorov[1]　(1. Universidade Federal de Viçosa(Brazil), 2. Handong GLobal University(Korea))

10:45 AM - 11:00 AM

[4-1015-C-04] **Sensitivity Comparison of Standard and real-time PCR Assay for Detection *Salmonella* Typhimurium and Enteritidis in Indonesian Chicken Carcasses**

*siti nurjanah[1,2], Winiati Puji Rahayu[1,2], Ratih Dewanti-Hariyadi[1,2]　(1. Department of Food Science &Technology, Bogor Agricultural University (IPB University)(Indonesia), 2. SEAFAST Center, Bogor Agricultural University (IPB University)(Indonesia))

11:00 AM - 11:15 AM

[4-1015-C-05] **Development of Calculation Framework for Stochastic Prediction of Uncertainty and Variability in Survival Spore Numbers during Non-isothermal Inactivation by Second-order Monte Carlo Simulation**

*Hiroki Abe[1], Kento Koyama[1], Kohei Takeoka[1], Shinya Doto[1], Shuso Kawamura[1], Shige Koseki[1]　(1. Hokkaido University(Japan))

11:15 AM - 11:30 AM

[4-1015-C-06] **Evaluation Growth Characteristics of Bacterial Spores Combine Treatment with High Hydrostatic Pressure and Alkaline Electrolyzed Water**

*Koki Morita[1], Taiga Kuhara[1], Yoshinori Kamitani[1], Daisuke Hamanaka[1]　(1. Kagoshima University faculty of agriculture(Japan))

11:30 AM - 11:45 AM

[4-1015-C-07] **Impact of Mechanization Development on Women and Hired Labor Utilizations of Small-Scale Rice Farming Operations in Kampar Region, Indonesia**

*UJANG PAMAN[1], Khairizal Kha, Hajry Arief Wahyudy　(1. RIAU ISLAMIC UNIVERSITY(Indonesia))

11:45 AM - 12:00 PM

Oral Session | Food Safety
[4-1015-C] Food Safety (1)
Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room C (3rd room)

[4-1015-C-01] Surface Pasteurisation of Fresh Chicken Meat using UV-C Technology

Arthur Jonathan Philip[1], Negah Nikanjam[1], Emilia Nowak[1], *Anthony Mutukumira[1] （1. Massey University(New Zealand)）

Keywords: Chicken, UV-C, Spoilage, Shelf-life

Fresh chicken meat is highly susceptible to surface contamination by spoilage and pathogenic microorganisms. The New Zealand safety standard stipulate that aerobic mesophilic counts (AMCs) present on surfaces of fresh chicken should be <7 log CFU.cm^{-2} by end of shelf-life when stored at 4°C. The study investigated the effect of continuous ultraviolet light at 254 nm (UV-C) on the surface of fresh chicken samples. To determine the optimum UV-C parameter, fresh chicken portions (skinless breast fillet, skinless thigh fillet, skin-on breast fillet, and skin-on thigh fillet) were treated with four UV-C dosages (50, 100, 200, and 300 mJ.cm^{-2}) at ambient temperature (20°C) using a commercial UV disinfection system. Temperature changes, exposure time and AMCs were determined as the responses. Standard enumeration of AMCs on fresh chicken portions was carried out by swabbing and plating dilutions on agar plate followed by 30°C/72 h incubation. The result indicated that 50 mJ.cm^{-2} UV-C dose had maximum microbial reduction on surfaces of skinless and skin-on chicken samples with minimal temperature changes and lowest exposure times hence, it was selected as the optimum dosage for the two types of fresh chicken samples. The effect of 50 mJ.cm^{-2} UV-C dose was then investigated on the surfaces of fresh skinless and skin-on chicken breast samples. Treated samples intended for storage at 4°C/7 days were repackaged. Instrumental color analysis, AMCs, lipid oxidation, and sensory evaluation were conducted during storage (4°C) for 7 days. All the chicken samples were also tested for *E. coli*, *S. aureus*, *L. monocytogenes*, *Campylobacter* spp. and *Salmonella* spp. The fresh chicken samples were evaluated for appearance, odor, and texture by 5 semi-trained focus group panelists at 1, 5, and 7 days. Cooked chicken samples were evaluated by consumer sensory panelists (n=30) on days 1 and 7 using the 9-point hedonic rating scale. AMCs on control chicken samples exceeded the national standard on day 5 of storage, whereas UV-treated chicken samples (p<0.05) extended the shelf life to day 6 (skin-on) and day 7 (skinless) by reducing the initial cell counts and 3.80 ± 0.35 log CFU.cm^{-2} skinless chicken samples to 3.07 ± 0.34 and 1.87 ± 0.98 log CFU.cm^{-2}, respectively. Insignificant (p>0.05) differences of microbial counts between the control and UV-treated skin-on samples were found. Tests for the pathogens were negative in all the chicken samples. Although 50 mJ.cm^{-2} UV-C dose had no impact (p>0.05) on the color and lipid oxidation of both skin-on and skinless chicken samples, the sensory panelists detected a slight burnt odor of UV-C treated fresh raw chicken samples stored (4°C) for day 1 which was not detected after cooking of the chicken samples that had been stored. The results suggested that 50 mJ.cm^{-2} continuous UV-C light (254 nm) chicken surface treatment successfully extended the shelf life by reducing the AMCs on skinless chicken portions compared to the skin-on samples.

Surface Pasteurisation of Fresh Chicken Meat using UV-C Technology

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ABSTRACT

Introduction

Fresh chicken meat is highly susceptible to surface contamination by spoilage and pathogenic microorganisms. The New Zealand safety standard stipulate that aerobic mesophilic counts (AMCs) present on surfaces of fresh chicken should be <6 log CFU cm^{-2} by end of shelf-life when stored at 4°C. The study investigated the effect of continuous ultraviolet light at 254 nm (UV-C) on the surface of fresh chicken samples.

Materials and Methods

To determine the optimum UV-C light dosage, fresh chicken portions (skinless breast fillet, skinless thigh fillet, skin-on breast fillet, and skin-on thigh fillet) were treated with four UV-C dosages (50, 100, 200, and 300 mJ cm^{-2}) at ambient temperature (20°C) using a commercial UV disinfection system. Temperature changes, exposure time and AMCs were determined as the responses. Standard enumeration of AMCs on fresh chicken portions was carried out by swabbing and plating dilutions on agar plate followed by 30°C/72 h incubation. The result indicated that 50 mJ cm^{-2} UV-C dose had maximum microbial reduction on surfaces of skinless and skin-on chicken samples with minimal temperature changes and lowest exposure times hence, it was selected as the optimum dosage for the two types of fresh chicken samples. The effect of 50 mJ cm^{-2} UV-C dose was then investigated on the surfaces of fresh skinless and skin-on chicken breast samples. Treated samples intended for storage at 4°C/7 days were repackaged. Instrumental color analysis, AMCs, lipid oxidation, and sensory evaluation were conducted during storage (4°C) for 7 days. All the chicken samples were also tested for *E. coli*, *S. aureus*, *L. monocytogenes*, *Campylobacter* spp. and *Salmonella* spp. The fresh chicken samples were evaluated for appearance, odor, and texture by 5 semi-trained focus group panelists at 1, 5, and 7 days. Cooked chicken samples were evaluated by consumer sensory panelists (n=90) on days 1 and 7 using the 9-point hedonic rating scale.

Results and Discussion

AMCs on control chicken samples exceeded the national standard on day 5 of storage, whereas UV-treated chicken samples (p<0.05) extended the shelf life to day 6 (skin-on) and day 7 (skinless) by reducing the initial cell counts of 3.31 ± 0.11 and 3.80 ± 0.35 log CFU.cm^{-2} to 3.07 ± 0.22 and 1.87 ± 0.98 log CFU cm^{-2} for skin-on and skinless chicken samples respectively. Insignificant (p>0.05) differences of microbial counts between the control and UV-treated skin-on samples were found, while significant differences (p<0.05) were found on the AMCs between control and UV-treated skinless chicken samples. Tests for the pathogens were negative in all the chicken samples. Although 50 mJ cm^{-2} UV-C dose had no impact (p>0.05) on the color and lipid oxidation of both skin-on and skinless chicken samples, the sensory panelists detected a slight burnt odor of UV-C treated fresh raw chicken samples stored (4°C) for day 1 which was not detected after cooking of the chicken samples that had been stored.

Conclusion and Recommendation

The results suggested that 50 mJ cm^{-2} continuous UV-C light (254 nm) chicken surface treatment successfully extended the shelf life by reducing the AMCs on skinless chicken portions compared to the skin-on samples.

Keywords: Chicken, UV-C, Spoilage, Shelf-life.

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1. INTRODUCTION

Fresh chicken meat is an important source of protein worldwide. In 2018, the annual world poultry production reached 123 million tons (McLeod et al., 2018). In New Zealand, the consumption of chicken meat is about 40 kg per person (PIANZ, 2018). Fresh chicken meat is highly susceptible to surface contamination by spoilage and pathogenic microorganism due to their rich nutritional content and high-water activity. Microorganisms are already present on the skin of the birds and gastrointestinal tract before slaughter. Therefore, there is a high possibility for these microorganisms to spread and contaminate the fresh meat product through cross-contamination during handling and processing. Food contamination and spoilage by microorganism present serious problems for consumer safety. There have been numerous cases of food poisoning, pathogen outbreak, and product recalls worldwide (Bardon, Kolar, Cekanova, Hejnar, & Koukalova, 2009; Castaneda, 2017; Premarathne et al., 2017; Scheinberg, Doores, & Cutter, 2013). *Campylobacter jejuni*, *Salmonella* spp., *Listeria monocytogenes*, and *Escherichia coli* are the main pathogenic microorganism found on the surface of chicken meat (Castaneda, 2017; Cunningham, 2012; Premarathne et al., 2017; Scheinberg et al., 2013). These bacteria can infect human through consumption of cross-contaminated or undercooked chicken meat (Devleesschauwer, Bouwknegt, Mangen, & Havelaar, 2017; Pasquali et al., 2017; Premarathne et al., 2017).

The presence of spoilage bacteria on the surface of fresh poultry meat after processing is the main reason in determining the shelf-life of the product (Octavian & Octavian, 2010; Petracci & Fletcher, 2002; Russell, Fletcher, & Cox, 1995). The growth of the spoilage microorganisms during cold-storage can lead to quality defects such as discoloration, development of off-odor, and development of off-flavor (James, 2005; Mead, 2004; Pearson & Dutson, 1994; Shall, 2013). Once the growth of the spoilage microorganisms reaches high numbers ($>10^5$ CFU cm^{-2}), they can produce metabolites that cause the defects to become more noticeable. The shelf-life of fresh poultry meat products ultimately end when the consumer is able to notice and identify these defects (Carvalho, Shimokomaki, & Estévez, 2017; Parrott & Walley, 2017). Therefore, international food safety authorities such as USFDA and FSANZ recommended that AMCs present on the surface of fresh chicken meat should be $<10^6$ CFU cm^{-2} at the end of shelf-life (FSANZ, 1995; Hasell & Salter, 2003).

Different food preservation methods have been developed and applied in food processing to control or reduce contamination thereby extending the shelf-life of food products. Heat and chemical treatment are the most common methods of microorganism inactivation (Huang, Wu, Lu, Shyu, & Wang, 2017; Tewari & Juneja, 2007). Despite being highly effective, heat treatment and chemical antimicrobial agent often destroy sensory properties and valuable nutrients such as protein and vitamins (Koutchma, 2008, 2009; Tewari & Juneja, 2007).

Alternative non-thermal and non-chemical preservation methods have been investigated for food processing application. Ultraviolet (UV) light technology, high pressure processing (HPP), high voltage processing, and gamma irradiation, are some of the examples of alternative preservation methods (Gould, 2001; Gunter-Ward et al., 2018; Lynch, 2016; Seemeen, 2011; Tewari & Juneja, 2007). UV light technology in particular, is a non-ionizing irradiation spectrum of light that possess germicidal capabilities (Ahmad, Christensen, & Baron, 2017; Gabriel, Ballesteros, Rosario, Tumlos, & Ramos, 2018; Gunter-Ward et al., 2018; Koutchma, 2008, 2009; Unluturk & Atilgan, 2014). UV light has several advantages over other alternative technologies including ease of operation and cost efficiency (Baysal, Molva, & Unluturk, 2013; Gunter-Ward et al., 2018).

The USFDA and European Food Safety Authority (EFSA) have recently approved the application of UV-C light (240-315 nm) for the treatment of various types of food such as bread, juice and dairy products (Forney & Moraru, 2009). UV-C was first applied as a substitute for heat treatment for the pasteurisation of liquid food products such as fruit juice and milk (Ahmad et al., 2017; Gabriel et al., 2018; Gunter-Ward et al., 2018). Successful applications of UV-C have also been reported for surface pasteurisation of fruit, vegetables, raw meat, and cooked meat (Baysal et al., 2013; Butot et al., 2018; Gamage, 2015; Heinrich, Zunabovic, Varzakas, Bergmair, & Kneifel, 2016; Semi, 2016)

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UV-C light technology has a huge potential for poultry processing application. The germicidal capabilities paired with minimal impact on sensory properties of the meat product make UV-C technology a suitable processing aid in the poultry industry. UV-C light processing can reduce the presence of spoilage bacteria on the surface of fresh chicken product directly after processing, resulting in the extension of shelf-life. Increasing the shelf life of fresh chicken meat product will lead to the increase in food safety, economic value, as well as decrease food waste. Despite the advantages of UV-C application in the food industry, application of the technology has been generally been slow worldwide, including in New Zealand (Koutchma, 2008, 2009). Therefore, the aim of this study was to investigate the potential of UV-C light processing for surface pasteurisation of fresh chicken meat portion.

2. MATERIALS AND METHODS
2.1 Samples
Skinless breast fillet (SLBF), skin-on breast fillet (SOBF), skinless thigh fillet (SOTF), and skin-on thigh fillet (SOT) chicken samples were supplied by a local commercial poultry processing factory in Auckland, New Zealand. The chicken portions were packed in bulk (5 kg) and delivered to the Microbiology Laboratory under chilled conditions (4°C) within one hour.

2.2. UV-C Light Surface Pasteurisation Optimisation
The goal of the optimisation was to select an optimum UV-C dosage for surface pasteurisation with maximum bacterial reduction of viable cell counts, minimum temperature increase, and lowest exposure duration. A factorial experiment consisting of 2 factors (UV-C light dosages and chicken meat type) with 4 levels was generated to determine the optimum dosage for continuous UV-C light surface pasteurisation process on fresh chicken meat portions.

2.3. UV-C light Surface Pasteurisation Treatment
The UV-C light surface pasteurisation was conducted using the Joulesafe® UV disinfection system (Radiant UV, USA). Twelve (12) low-pressure mercury lamps, each emitting 254 nm UV light were the sources of the UV-C light. The mercury lamps were mounted at the top and bottom of the UV-C chamber, providing complete coverage of UV light exposure. The equipment also recorded the intensity and exposure duration of each cycles of UV-C light exposure process. The UV-C light surface pasteurisation dosage was automatically adjusted by the equipment. The fresh chicken samples were treated with four different UV-C dosages: 50, 100, 200, and 300 mJ cm^{-2} at ambient temperature (20°C). The samples were placed on a custom-made tray provided by the manufacturer and placed inside the equipment for UV-C light exposure process. Following UV-C light exposure, the chicken samples were individually packed using polyethylene terephthalate (PET) tray with a polyethylene sealant layer and stored for 7 days at refrigeration temperature (4°C).

The temperature on the surface of the chicken samples was measured using an RS PRO RS-41 thermometer (RS Components Ltd., UK) equipped with a 20-cm probe before and after UV-C treatment. Temperature measurement were conducted in triplicates.

2.4. Microbiological analysis
Wet and dry swab sampling procedure of ISO 18593 was used to collect swab samples (Castaneda, 2017). Following UV-C light surface pasteurisation, the surface of each sample was swabbed with three sterile cotton swabs (Nanjing Luster Medical & Healthcare Products, China) and a 5-cm^2 sterile template (Fort Richard, NZ). Firstly, the first swab was pre-moistened using maximum recovery diluent (MRD) (Oxoid, NZ) at 0.1% (w/w). The pre-moistened swabs were used to swab the surface of the chicken samples for 30 seconds. The tips of the swabs were aseptically broken and placed into 10-ml sterile peptone water. The second and third swabs were used to swab the same surface to ensure maximum recovery of bacteria from the surface of the sample. The glass bottle containing the swab samples and MRD were mixed thoroughly using a vortex mixer (VM-96B JEIO TECH, Korea). Serial dilutions of the samples were prepared up to 10^{-6} dilution. Each dilution was aseptically transferred (0.1 mL) to the surface of suitable medium (solidified agar) for each respective bacterial species (except for *Escherichia coli)*. Agar plates for aerobic mesophilic counts (AMC) were incubated at

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30°C/ 72 h; *Salmonella* spp., *Staphylococcus aureus,* and *Listeria Monocytogenes* were incubated at 35°C/24 h. For *Campylobacter* spp., the mCCDA plates were stacked inside an airtight rectangular plastic container (Castaneda, 2017). Two CampyGen™ (ThermoFischer Scientific, USA) sachets were placed inside the container to create microaerophilic conditions (84% N_2, 10% CO_2, and 6% O_2). The plates were then incubated at 42°C for 48 h. Developed colonies were counted after incubation and expressed as log_{10} CFU cm^{-2}.

The enumeration of *Escherichia coli* was carried out using the pour plate method. VRB agar was prepared following the manufacturer's instruction on the day of experiment and was kept at 48°C (Castaneda, 2017; Chun, Kim, Lee, Yu, & Song, 2010). One ml of each dilution was pour plated on the VRB agar. The solidified plates were incubated at 35°C/24 h. After incubation, developed colonies were enumerated as previously described.

2.5. Effect of UV-C Light Surface Pasteurisation during Storage
Following optimisation, one optimum UV-C light dosage was selected. The effect of the optimum dosage on the psychochemical and sensory quality of fresh chicken meat portions during refrigerated storage (4°C) was analysed by conducting microbial enumeration, color measurement, lipid oxidation analysis, and sensory evaluation on UV-C treated and untreated fresh chicken samples (SLBF and SOBF) during 7-day storage under refrigerated conditions (4°C). Microbial enumeration was conducted as described in the previous sub-chapter at day 0, 3, 5, and 7 of storage.

2.5. Lipid Oxidation Analysis and Instrumental Color Measurement
The degree of lipid oxidation of the untreated and UV-C treated fresh chicken samples was measured using the TBA method described by Chun et al. (2010) and Ahn et al. (1998). Five grams of sample were homogenized using a blender (BFP 100, Breville Inc, Australia) in 15 mL distilled water. One mL of each meat (sample) homogenate and 2 mL of 20 mM 2-thiobarbituric acid/15% trichloroacetic acid (TBA/TCA) solution were transferred into a test tube. The mixture was mixed using a vortex mixer (VM-96B JEIO TECH, Korea) and incubated in boiling water for 15 minutes to develop color. The mixture allowed to cool to room temperature (20°C) and then centrifuged (6-16KS, Sigma, Germany) at 2000 *g* for 15 minutes. The absorbance of the resulting supernatant was measured using a spectrophotometer (UV-1601, Shimadzu, Japan) at 531 nm. Thiobarbituric acid reactive substances (TBARS) values represented the degree of lipid oxidation and were calculated using the malonaldehyde standard curve prepared using 1,1,3,3-tetra-ethoxypropane (TEP). The TBARS values were expressed as milligrams malonaldehyde in one kilogram of meat sample (MDA/kg). The lipid oxidation measurement was conducted in three replicates.

A Minolta CR-300 Colorimeter (Minolta Company, Tokyo, Japan) equipped with illuminant D65 as the light source was used to measure the color of the samples (Chun et al., 2010; Seemeen, 2011). The measuring head of the CR-300 used diffuse illumination/0° viewing geometry to measure the color of the samples as viewed under diffuse lighting conditions. The colorimeter was calibrated using a standard white calibration plate provided by the manufacturer with L* value of 94.01, a* value of 0.29, and b* value of 1.77. The color measurements were conducted in five replicates. The Hunter's color values (L*, a*, b*) and lipid oxidation of the samples were measured and recorded on days 0, 3, 5, and 7 during storage.

2.7. Sensory Evaluation
The sensory evaluations of untreated and UV-treated chicken meat samples were conducted by a focus group and consumer sensory participants who used the 9-point hedonic scale (McLeod et al., 2018; Seemeen, 2011). A focus group comprising of five panelists evaluated the raw chicken samples during storage for seven days. Four samples (untreated skin-on chicken breast, UV treated skin-on chicken breast, untreated skinless chicken breast, and UV treated skinless chicken breast) were presented to the focus group panelists. The panelists evaluated the samples for appearance, texture and odor. Ninety (90) consumer sensory evaluation participants evaluated the cooked samples (cooked untreated and UV-C treated chicken) using the 9-point hedonic scale at days 1 and 7 of storage (McLeod et al., 2018; Seemeen, 2011). The chicken samples were roasted in a convection oven (Bakbar Turbofan 32 Max,

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Moffat, Australia) without any seasoning to a center temperature of 75°C. Internal temperature of the samples during cooking process was monitored using a temperature probe (Part number – 16002, Sensing Devices Limited, UK) connected to a probe log. The cooked products were cooled to 30 – 40°C and then cut into uniform portions (2 x 2 cm) for sensory evaluation. Four different cooked chicken samples (untreated skinless breast, UV-treated skinless breast, untreated skin-on breast, and UV-treated skin-on breast) were evaluated by sensory panelists. The samples from each treatment were coded with random three-digit numbers before being presented to the sensory panelists. Sensory panelists were provided with crackers and still bottled water to rinse their palates during sensory evaluation. The consumer sensory panelists evaluated the test samples for taste, texture, flavor, freshness and the overall acceptability of the sample on a 9-point hedonic scale.

2.8. Statistical Analysis
The data obtained in this study were analysed using Minitab Statistical Software version 17.0 (Minitab Inc., USA, 2009). Data on optimisation/selection of UV-C light surface pasteurisation dosage was analysed using the general linear model. Microbial cell counts, TBARS, Hunter L*, a*, b* values, and sensory evaluation data obtained during storage were analysed using one-way analysis of variance (ANOVA) and significant means were separated by Tukey's post-hoc test.

3. RESULTS AND DISCUSSION
3.1 Selection of Optimum UV-C Light Surface Pasteurisation Dosage
The goal of the optimisation of UV-C light surface pasteurisation was to select a dosage capable of maximum reduction of AMCs with minimum temperature increase and exposure duration. The AMCs reduction of UV-C light dosages at 50 to 300 mJ cm^{-2} ranged from 1.69 to 2.98 log CFU cm^{-2} and 0.21 to 0.86 log CFU cm^{-2} for skinless and skin-on samples, respectively. The result indicated that increasing UV-C light dosages from 50 to 300 mJ cm^{-2} had no significant effect on the microbial reduction ($p>0.05$). The results also suggested that the reduction of AMCs was more effective ($p<0.05$) on the surface microflora of skinless chicken meat samples compared to their skin-on counterparts, which is in agreement with studies conducted by Isohanni and Lyhs (2009) and Haughton et al. (2011). UV-C light treatment on the surface of fresh chicken meat portions using dosages of 50 – 200 mJ cm^{-2} reduced AMCs from 1.41 to 1.76 log CFU cm^{-2} and 0.05 to 0.14 log CFU cm^{-2} for skinless and skin-on chicken breast fillet respectively (Haughton et al., 2011; Isohanni & Lyhs, 2009). The previous studies also concluded that increasing the UV-C light dosage from 50 up to 200 mJ cm^{-2} did not increase microbial reduction on both skinless and skin-on chicken meat samples ($p<0.05$).

Absorption of UV-C light by the DNA causes mutation in the microorganisms leading to cell death (Gabriel et al., 2018; Heinrich et al., 2016; Koutchma, 2008). As the method relies on direct exposure of light on the microorganisms residing on the surface, factors such as surface topography, UV-C light dosage and equipment, affect the inactivation efficiency (Chun et al., 2010; Gayán, Álvarez, & Condón, 2013; Heinrich et al., 2016). The surface of chicken skin is highly irregular in texture, which shelters microorganism from UV-C light (Haughton et al., 2011; Petracci & Fletcher, 2002). The surface topography of chicken meat is smoother than the skin, therefore the UV-C light treatment is more effective on skinless chicken meat surfaces.

The result also suggested that temperature increase and exposure duration were mainly influenced by the UV-C light dosage. Temperature increased from 2.43 to 15.77°C and 2.93 to 12.83°C for skinless and skin-on chicken samples respectively, when treated with UV-C dosage of 50 to 300 mJ cm^{-2}. There was no difference ($p<0.05$) between the temperature increases on skinless and skin-on chicken samples treated with the same UV-C light dosages. The duration of UV exposure ranged from 2.16 to 12.58 minutes (skinless) and, 2.16 to 13.03 minutes (skin-on) for samples treated with 50 to 300 mJ cm^{-2}. No significant differences between the exposure duration on skinless and skin-on chicken samples treated with the same UV-C light dosage ($p<0.05$). Prolonged UV-C exposure on the surface of chicken product can generate heat which may accelerate lipid oxidation, produce off-flavors and off-odors (Chun et al., 2010; Haughton et al., 2011; McLeod et al., 2018; Park & Ha, 2015). Therefore, it is important to select optimum UV-C dosages with low exposure times to minimize the increase in

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temperature during the surface pasteurisation process. In this study, the UV-C light dosage of 50 mJ cm^{-2} was selected as the optimum dosage since it possesses bacterial reduction capabilities that is not significantly different (p>0.05) compared to other dosages (100, 200, and 300 mJ cm^{-2}) while demonstrating the lowest exposure duration and minimal temperature increase.

3.2 Effect of Optimum UV-C Light Dosage on the Growth of Bacteria

The effect of selected (optimum) UV-C light surface pasteurisation dosage (50 mJ cm^{-2}) on the growth of microorganism (AMCs) during refrigerated storage (4°C) is shown in Figure 1. The initial AMCs present on the surface of the fresh chicken meat were 3.31 ± 0.11 and 3.80 ± 0.35 log CFU cm^{-2} for skin-on and skinless chicken meat samples, respectively. The initial AMCs were reduced to 3.07 ± 0.22 and 1.87 ± 0.98 log CFU cm^{-2} giving microbial log reduction value of 0.24 and 1.93, respectively. Thus, the microbial reduction on skin-on chicken breast fillet samples after UV exposure was insignificant (p>0.05), whereas significant AMCs reduction were observed on skinless chicken breast fillet (p<0.05). During storage trials, the two types of products (skin-on and skinless breast fillet) treated with UV-C light showed delayed AMCs growth compared to untreated (control) samples and the effectiveness was more pronounced towards the end of storage time. After the initial reductions, the differences in the microbial counts were stable (p<0.05) during storage. The AMCs for untreated skinless chicken breast surpassed the recommended limit set by FSANZ (1995) after day 5 of storage, while the AMCs for UV-C treated skinless breast surpassed the limit after day 7 of storage (Figure 1A). The AMCs for untreated skin-on chicken breast surpassed the limit day 5 of storage, while the AMCs for UV-treated skin-on chicken breast was higher than the limit on day 6 of storage (Figure 1B).

There are limited studies on the effectiveness of UV-C treatment on the growth of pathogens on the surface fresh chicken meat during refrigerated storage (4°C). Chun et al. (2010) showed the effectiveness of 50 mJ cm^{-2} UV-C light on growth of surrogate pathogens (*C. jejuni, L. monocytogenes, Salmonella* Typhimurium) on skinless chicken breast fillet during storage for 6 days at 4°C. In the present study, *Campylobacter* spp., *Salmonella* spp., *E. coli, S. aureus,* and *L. monocytogenes* were not detected in any of the samples which indicated effectiveness of the immersion chilling step in the commercial poultry processing factory (Demirok et al., 2013; James, Vincent, de Andrade Lima, & James, 2006; Zhang, Jeong, Janardhanan, Ryser, & Kang, 2011).
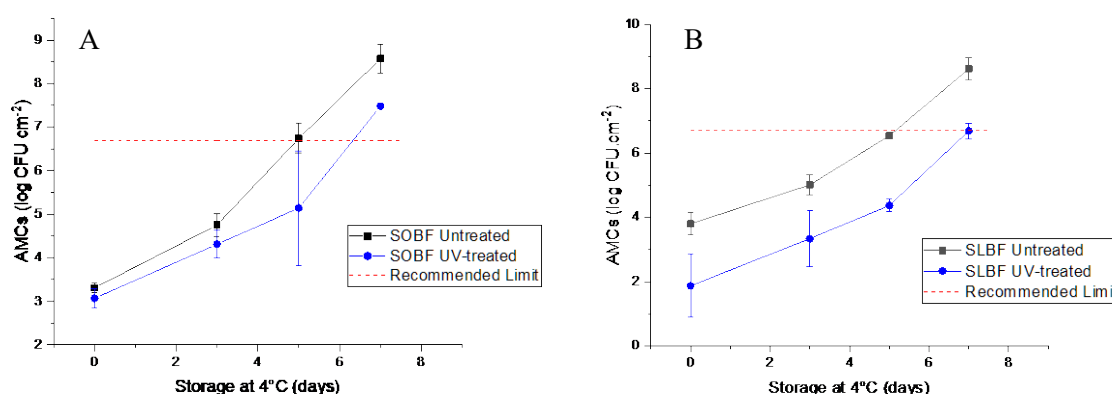

Figure 1. Aerobic mesophilic counts (AMCs) of untreated and UV-C treated (50 mJ cm^{-2}) fresh chicken meat samples.

Notes: SLBF = Skinless Breast Fillet (A); SOBF = Skin-on Breast Fillet (B); Dashed horizontal line = FSANZ (1995) microbiological criteria for foods recommended AMCs ($<5 \times 10^6$ CFU cm^{-2}) on fresh chicken meat product at the end of shelf-life. Error bars represent the standard deviation of the mean values; experiment was conducted in 2 replicates with 3 determinations each.

3.3 Color and Lipid Oxidation

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Lipid oxidation and color are two of the most important non-microbial physicochemical attributes of fresh chicken meat product (Carvalho, Shimokomaki, & Estévez, 2017). It was therefore important to evaluate impact of UV-C light processing on the two physicochemical parameters on fresh chicken meat samples during refrigerated storage (4°C). The degree of lipid oxidation (shown as TBARS values) is shown in Table 1. The changes of the TBARS in both UV-treated and untreated skin-on and skinless chicken breast were not significant (p>0.05). The difference of TBARS value between UV treated and untreated sample as well as the changes of TBARS value throughout the storage period was not significant (p>0.05).

Table 1. TBARS (MDA/kg) * of chicken breast fillet samples during storage at 4°C.

Sample Type	Storage (day)	Untreated (Control)	UV-C Light Treated (50 mJ cm^{-2})
Skin-on breast fillet	0	[a]1.72 ± 0.56[a]	[a]3.09 ± 0.70[a]
	3	[a]1.24 ± 0.36[a]	[a]2.14 ± 1.38[a]
	5	[a]1.40 ± 0.08[a]	[a]2.22 ± 0.27[a]
	7	[a]3.23 ± 1.39[a]	[a]2.46 ± 0.83[a]
Skinless breast fillet	0	[a]2.11 ± 0.75[a]	[a]1.34 ± 0.89[a]
	3	[b]0.58 ± 0.14[a]	[a]0.22 ± 0.21[a]
	5	[ab]1.03 ± 0.06[a]	[a]1.61± 0.64[a]
	7	[a]2.14 ± 0.35[a]	[a]1.43 ± 0.65[a]

Notes: Means* of TBARS followed by standard deviation (±). Within columns of each sample type, different superscripts in front of mean values indicate significance (p<0.05). Within rows of each sample type, mean values followed by different superscript letters are significantly different (p<0.05); MDA = Malonaldehyde. Experiment was conducted in two replicates with three determinations each.

The color measurement results indicated that UV-C light treatment had a significant effect (p<0.05) on the Hunter L*, a*, and b* values of skin-on chicken breast fillet during storage (Table 2). UV-C light treatment had a slight effect on the b* values and no effect (p>0.05) on the Hunter L* and a* values of skinless chicken breast fillet during storage. The slight changes of b* values have been previously reported (Chun et al., 2010; Park & Ha, 2015). Overall, UV-C light treatment had more impact on the physicochemical attributes of skin-on chicken samples compared to skinless chicken samples. Published data reported that UV-C light treatment have no significant effect on the physicochemical attributes of skinless chicken meat during storage. Chun et al. (2010), Park and Ha (2015), and McLeod et al. (2018) all reported that UV-C light treatment at 50 mJ cm^{-2} have no significant effect on the rate of lipid oxidation and color of skinless chicken breast fillet.

Table 2. Hunter L*, a*, and b* values* of chicken breast fillet samples during storage at 4°C.

Sample Type	Hunter values	UV-C dosage	Storage period (day)			
			0	3	5	7
Skin-on breast fillet	L*	Control	60.81 ± 0.33[a]	56.80 ± 0.01[d]	57.66 ± 0.01[c]	57.66 ± 0.01[c]
		50 mJ cm^{-2}	60.68 ± 0.04[a]	58.61 ± 0.01[b]	56.34 ± 0.02[e]	56.34 ± 0.02[e]
	a*	Control	-1.12 ± 0.01[a]	-1.05 ± 0.01[b]	-0.96 ± 0.01[c]	-0.53 ± 0.01[f]
		50 mJ cm^{-2}	-1.06 ± 0.01[b]	-0.96 ± 0.01[c]	-0.86 ± 0.01[d]	-0.58 ± 0.01[e]
	b*	Control	0.97 ± 0.01[e]	0.78 ± 0.03[f]	0.63 ± 0.02[g]	0.41 ± 0.02[h]
		50 mJ cm^{-2}	1.78 ± 0.05[a]	1.58 ± 0.02[b]	1.44 ± 0.02[c]	1.16 ± 0.01[d]
Skinless breast fillet	L*	Control	42.37 ± 0.09[a]	41.35 ± 0.13[b]	40.78 ± 0.37[c]	40.24 ± 0.01[d]
		50 mJ cm^{-2}	42.39 ± 0.01[a]	41.21 ± 0.01[b]	40.63 ± 0.03[c]	40.24 ± 0.01[d]
	a*	Control	1.14 ± 0.02[a]	1.09 ± 0.01[b]	0.74 ± 0.01[c]	0.61 ± 0.03[d]
		50 mJ cm^{-2}	1.16 ± 0.01[a]	1.08 ± 0.01[b]	0.73 ± 0.01[c]	0.62 ± 0.01[d]
	b*	Control	1.30 ± 0.02[a]	0.83 ± 0.03[c]	0.63 ± 0.04[d]	0.41 ± 0.01[e]
		50 mJ cm^{-2}	0.92 ± 0.03[b]	0.83 ± 0.01[c]	0.59 ± 0.02[d]	0.46 ± 0.02[e]

Notes: Means* of Hunter L*, a*, and b* values followed by standard deviation (±). L*, degree of lightness (0 – 100 = black – white); a*, degree of redness ((-80) – (+100) = green – red); b*, degree of yellowness ((-80) – (+70) = blue – yellow). Within columns and rows of each sample type and color values (L*, a*, and b*), mean values followed by different superscript are significantly different (p<0.05). Experiment was conducted in two replicates with five determinations each.

3.4 Sensory Evaluation

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Raw untreated and UV-C light treated (50 mJ cm^{-2}) skinless and skin-on chicken breast samples were evaluated for appearance, odor, and texture by a focus group on day 1, 5, and 7 of storage. The focus group found no difference in the appearance and texture throughout the storage time, however burnt odor on UV-C light treated chicken samples was detected on day 1 of storage. Similar off-odor on the chicken samples immediately after UV-C treatment was previously reported by McLeod et al. (2018). Further sensory evaluation during storage by the focus group revealed that the burnt odor was no longer noticeable on samples stored for 5 and 7 days.

Sensory evaluation was conducted on cooked untreated (control) and UV-C treated chicken meat samples using hedonic test with 90 participants. Cooking the chicken samples was done to mimic normal practices in chicken consumption. The result of hedonic test showed that UV-C light surface pasteurisation had no impact ($p > 0.05$) on the appearance, flavor, texture, juiciness, and overall acceptance of both cooked skinless and skin-on chicken samples throughout refrigerated storage (data not shown). The burnt odor detected on day 1 of storage by the focus group on raw UV-C treated chicken samples was not detected on the cooked product and similar impact of cooking on odor fading was reported by McLeod et al. (2018). Similar work done by Park & Ha (2015) showed no effect on sensory attributes of UV-C treated (60 mJ cm^{-2}) skinless chicken breast samples. The results of sensory evaluation in this study showed that UV-C light surface pasteurisation had no impact on the sensory attributes of fresh chicken meat stored for seven days at (4°C).

4. CONCLUSION
The result of this study suggested that UV-C light surface pasteurisation dosage of 50 mJ cm^{-2} has the potential to extend the shelf-life of fresh skinless chicken meat portions without significant impact on both sensorial and physicochemical attributes. The result also suggested that UV-C light processing was not effective for the surface pasteurisation of fresh skin-on chicken meat portions.

ACKNOWLEDGMENT

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10:30 AM - 10:45 AM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room C)

[4-1015-C-02] Efficient Filtering of Live *Escherichia coli* by Using 60 GHz CMOS Sensor

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Keywords: Bacteria separation, 60 GHz CMOS sensor, Dielectrophoresis, E. coli

The separation and extraction of bacteria are essential for food safety evaluation and infection diagnosis by a blood test. In particular, the quick selection of live and dead bacteria can contribute to developing efficient and high precision bacterial testing. To realize this, we have been developing a near-field array CMOS sensor with a 60 GHz oscillator. 1488 oscillators are arranged in 3 mm square, and when the sample exists on the surface, the distribution of the electric field on the sensor surface changes and the resonance frequency also changes. We succeeded in mounting the circuit of dielectrophoresis (DEP) in each oscillator by CMOS integration technology. In this presentation, we report the investigation of the optimization of applied voltage and frequency to this DEP circuit in order to distinguish live and dead bacteria.

 The cultured *E. coli* suspension was centrifuged twice to replace the medium and distilled water. This *E. coli* suspension was used as a measurement sample of viable bacteria. Also, 5 ml of this suspension was placed in an autoclave for 1 hour to make dead bacteria. And the oscillation frequency of two samples of live and dead bacteria was measured every 6 seconds for 300 seconds. In the measurement, the sample was flowed at 5 μ L / min with a syringe pump (YWC). In addition, DEP was set to a voltage of 5 V and a frequency of 10, 100, and 1000 kHz. DEP was turned off from 0 to 60 seconds, DEP was turned on from 60 to 240 seconds, and DEP was turned off from 240 to 300 seconds. The experiment was conducted while confirming the presence of *E. coli* on the CMOS sensor with a microscope.

 The oscillation frequency at 240 seconds minus the oscillation frequency at 300 seconds was taken as the frequency shift. At DEP 10 kHz, it is thought that no live bacteria or dead bacteria were affected by the negative DEP force and bacteria did not accumulate on the oscillator. At DEP 100 kHz, a positive force of DEP acts on the live bacteria, and the live bacteria are accumulated on the oscillator, and the shift of the live bacteria is considered to be 0.015 GHz. It is thought that bacteria did not accumulate on the oscillator because negative DEP force was exerted on dead bacteria. At DEP 1000 kHz, it is thought that positive DEP force acts on both live and dead bacteria, and bacteria accumulate on the oscillator. From this experiment, the possibility of distinction between live and dead bacteria of *E. coli* was shown at DEP 100 kHz.

Oral Session | Food Safety
[4-1015-C] Food Safety (1)
Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room C (3rd room)

[4-1015-C-03] Safety Evaluation of Bacteriocinogenic Strains of *Pediococcus acidilactici* Isolated From Artisanal Cheeses

Luis Augusto Nero[1], Yosep Ji[2], Wilhelm Holzapfel[2], *Svetoslav Dimitrov Todorov[1] （1. Universidade Federal de Viçosa(Brazil), 2. Handong GLobal University(Korea)）
Keywords: Pediococcus acidilactici, bacteriocins, virulence factors

Total DNA extracted from bacteriocinogenic *Pediococcus acidilactici* ST1607V, ST2104V and ST3105V, was been screened for presence of more than 50 genes related to production of biogenic amines (histidine decarboxylase, tyrosine decarboxylase and ornithine decarboxylase), virulence factors (sex pheromones, gelatinase, cytolisin, hyaluronidase, aggregation substance, enterococcal surface protein, endocarditis antigen, adhesion of collagen, integration factors) and antibiotic resistance (vancomycin, tetracycline, erythromycin, gentamicin, chloramphenicol, bacitracin). *Pediococcus acidilactici* ST1607V, ST2104V and ST3105V presented low frequencies of presence for virulence genes. Only few genes were detected in some strains, indicating their safety for application in fermented food products. Besides all beneficial properties studied for various LAB, most considered as GRAS, a special attention need to be addressed on the possible presence of virulence factors, production of biogenic amines and antibiotic resistance. Results from appropriate biochemical tests and detected main genes associated to virulence factors and antibiotic resistance in LAB strains, could be considered as potential hazard of application of this organisms in food products. Moreover, additional *in vitro* and *in vivo* experiments must be designed in the system simulating fermentation processes or GIT conditions in order to investigate deeper the possible expression of the present virulence genes.

Safety Evaluation of Bacteriocinogenic Strains of *Pediococcus acidilactici* Isolated From Artisanal Cheeses

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ABSTRACT

Total DNA extracted from bacteriocinogenic *Pediococcus acidilactici* strains ST1607V, ST2104V and ST3105V, was screened for the presence of more than 50 genes related to the production of biogenic amines (histidine decarboxylase, tyrosine decarboxylase and ornithine decarboxylase), virulence factors (sex pheromones, gelatinase, cytolysin, hyaluronidase, aggregation substance, enterococcal surface protein, endocarditis antigen, adhesion of collagen, integration factors) and antibiotic resistance (vancomycin, tetracycline, erythromycin, gentamicin, chloramphenicol, bacitracin). *Pediococcus acidilactici* ST1607V, ST2104V and ST3105V presented low frequencies of presence for virulence genes. Only a few genes related to safety were detected in some strains, indicating their safety for application in fermented food products. In addition to the beneficial properties widely studied and reported for various lactic acid bacteria (LAB), most are considered as GRAS generally recognized as safe) organisms. However, special attention should be given to the presence of possible virulence factors, in addition to the production of biogenic amines and antibiotic resistance. Results from appropriate biochemical tests and based on major genes associated with virulence factors and antibiotic resistance, may serve to identify a potential hazard before application in food products. Moreover, additional *in vitro* and *in vivo* experiments could be designed for simulating fermentation processes and/or GIT conditions in order to determine possible expression of detected virulence genes.

Keywords: *Pediococcus acidilactici,* Bacteriocins, Virulence factors

1. INTRODUCTION

Application of LAB is well accepted in different areas of bio-preservation of food products such as starter or adjunct cultures for control of spoilage microorganisms. Traditionally, LAB have a long history of safe use and association with fermentations of food products from plant, meat and dairy origin. On the basis of empiric knowledge appropriate safe cultures are being isolated and selected for appropriate applications. Different LAB strains are part of numerous fermented food products from different geographical regions. However, even if numerous studies report on their beneficial properties (probiotics, starter and biopreservation cultures), only a limited number of studies have focused thus far on the safety of these strains. The majority of the LAB strains, especially of *Lactobacillus* spp., *Pediococcus* spp. and *Lactococcus* spp., have been Generally Recognized as Safe (GRAS) for human and animal applications based on their historical safety record and applications in different food fermentations. However, scanning the existing literature it is scarcely realized that some clinical cases of infection were linked to some LAB strains (Goldstein et al., 2015).

One of the essential factors in evaluation of safety of beneficial LAB in the future is the presence of virulence factors in addition to (transferable) antibiotic resistance. Absence of genes related to virulence factors has become a most important criterion in the safety evaluation of LAB. Beneficial LAB carrying virulence genes, especially of easy transferable genetic material, can be involved in a spreading of these genes via horizontal transfer to other bacteria, including to some relevant human and animal pathogens. It is thus imperative to know the potential virulence related gene profile of such beneficial LAB strains, and thereby prevent them serving as potential reservoirs of resistance genes that can result in th generation of potentially hazardous strains (Todorov et al., 2019).

In this work we explore safety aspects of 3 bacteriocinogenic strains of *Pediococcus acidilactici* isolated from artisanal cheese in Brazil, related to the presence of genes associated to virulence, antibiotic resistance and production of biogenic amines in addition to basic antimicrobial properties.

2. MATERIALS AND METHODS

2.1. Isolation of bacteriocin producers from cheese

Screening for bacteriocin-producing isolates was carried out according to the triple-agar-layer method described by Todorov & Dicks (2005). Samples of 50 g each of artisanal cheeses obtained from the local market (Vicosa, MG, Brazil), were macerated with 450 ml sterile saline (0.85%, w/v NaCl) in a Stomacher (BagMixer, Interscience, Weymouth, USA) for 10 min at 20 °C. Serial dilutions of the sample were made with sterile saline, plated onto MRS agar (Difco). The second layer of agar (1.0 %, w/v) was supplemented with 50.0 mg/L Actidion (Sigma) to prevent fungal growth. All plates were incubated at 37 °C for 24 h. Colonies were then overlaid with a third layer of 1% (w/v) Brain Heart Infusion (BHI) agar (Difco), seeded with 10^6 CFU/mL of *Listeria monocytogenes* 711. The plates were incubated at 37 °C for 24 h. Colonies with the largest zones of growth inhibition were isolated, inoculated into MRS broth (Difco) and incubated at 30 °C for 24 h. Pure cultures were obtained by streaking onto MRS agar.

Antimicrobial activity was confirmed by using the agar-spot-test method (Todorov, 2008). Activity was expressed as arbitrary units (AU) per mL, with one AU defined as the highest dilution showing a clear zone of inhibition (Todorov, 2008). *L. monocytogenes* 711 was used as a sensitive test strain.

One mL of a cell-free supernatant, prepared as described before, was added to 1 mg/mL of α-amylase (Sigma Diagnostics, St. Louis, MO, USA), Proteinase K (Roche, Indianopolis, IN, USA) and pronase, lipase or catalase (Roche), respectively. Samples were incubated at 30 °C for 30 min and then heated at 95–97 °C for 5 min. The pH of all samples was adjusted to 6.0 and bacteriocin activity determined with *L. monocytogenes* 711 as sensitive strain, as described above.

2.2. Identification of strains

Identification was conducted by physiological and biochemical tests (Stiles & Holzapfel, 1997). Carbohydrate fermentation reactions were recorded by using API 50 CHL (Biomérieux, Marcy-I'Etiole, France). Total DNA from the selected strains was isolated using the ZR Fungal/Bacterial DNA Kit (Zymo Research, Irvine, CA, USA) following the instructions of the manufacturer. DNA concentration was determined on NanoDrop (ThermoFisher Scientific Inc., Waltham, MA, USA). Differentiation of the strains was performed by random amplification of polymorphic DNA (RAPD) PCR. Primers OPL-02 and OPL-08 were used (Kit L of the RAPD® lomer kits, Operon Biotechnologies, Cologne, Germany). Amplification reactions were performed according to Todorov et al. (2010). The amplified products were separated by electrophoresis in 1.4% (w/v) agarose gels in 1x TAE buffer at 100V for 2 h. Gels were stained in TAE buffer containing 0.5 μg/mL of ethidium bromide (Sigma Diagnostics, St. Louis, Mo., USA).

Identification of the studied strains was performed by amplifying the genomic DNA with primers F8 (5'-CAC GGA TCC AGA CTT TGA TYM TGG CTC AG-3') and R1512 (5'-GTG AAG CTT ACG GYT AGC TTG TTA CGA CTT-3'), as described by Felske et al. (1997). The amplified fragments were cleaned using SigmaSpin™ Post-Reaction Clean-Up Columns (Sigma, St Louis, MO, USA), sequenced, and compared to sequences in GenBank using BLAST (Basic Local Alignment Search Tool).

2.3. Detection of virulence genes:

All PCR reactions were performed using the GeneAmp® PCR Instrument System 9700 (Applied Biosystems, Foster City, USA). All strains were tested for presence of virulence genes: production of gelatinase (*gel*E), hyaluronidase (*hyl*), aggregation substance (*asa*1), enterococcal surface protein (*esp*), cytolysin (*cyl*A), endocarditis antigen (*efa*A), adhesion of collagen (*ace*), vancomycin resistance (*van*A, *van*B, *van*C1, *van*C2, *van*C2/C3), erythromycin resistance (*erm*A, *erm*B, *erm*C), tetracycline resistance (*tet*K, *tet*L, *tet*M, *tet*O, *tet*S), related to gentamycin resistance (*aac*(6')-Ie-*aph*(2")-Ia), aminoglycosides resistance (*aph*(3')-IIIa, *ant*(4')-Ia, *aph*(2'')-Id, *aph*(2'')-Ic, *aph*(2'')-Ib, *ant*(6)-Ia), chloramphenicol resistance (*cat*A), bacitracin resistance (*bcr*B, *bcr*D, *bcr*R), production of sex pheromones (*ccf*, *cob*, *cpd*), serine protease (*spr*E), transposon related (*int*, *int*Tn) and genes for amino acid decarboxylases: *hdc*1 and *hdc*2 (both related to histidine decarboxylase), *tdc* (tyrosine

decarboxylase), and *odc* (ornithine decarboxylase), using PCR protocols of Moraes et al. (2012) and Fortuna et al. (2008).

3. RESULTS AND DISCUSSION

Based on inhibition zones of *L. monocytogenes* 711, 17 isolates were selected for future study. The selected isolates showed strong inhibitory activity against 30 different *L. monocytogenes* strains.

RAPD-PCR analysis with primers OPL-02 and OPL-08 (data not shown) showed differences among the 17 isolates and clearly established 3 distinct clusters. According to these results the 3 isolates (one from each cluster: ST1607V, ST2104V and ST3105V) were selected for future study. The 16S rRNA amplified from selected 3 strains revealed 98-99% homology to the 16S rRNA sequence of *P. acidilactici*.

The bacteriocins ST1607V, ST2104V and ST3105V were inactivated by treatment with the tested proteolytic enzymes, but not with α-amylase, lipase or catalase. This suggested that the activity of bacteriocins ST1607V, ST2104V and ST3105V was a direct effect of an antimicrobial proteinaceous metabolite.

The bio-molecular screening for presence of 50 virulence genes showed that *P. acidilactici* strains ST1607V, ST2104V and ST3105V carried at least a few virulence genes. However, no evidence for the presence of *asa1, aac(6')-Ie-aph(2")-Ia, aph(2")-Ic, aph(2")-Id, hdc1, hdc2, tdc, ddlE, bcrD, catA, ant(6)-Ia, aph(2")-Ib, aph(3')-IIIa, ace, cob, cpd, cyt, ermA, gelE, tetK, tetM, tetS, vanB, int* in any of the 3 tested strains was recorded (Table 1). Moreover, the observed frequency of putative virulence factors in *P. acidilactici* strains ST1607V, ST2104V and ST3105V was lower than that reported in other studies on *Lactobacillus* spp., *Enterococcus* spp., *Pediococcus* spp. isolated from foods (Todorov et al., 2014; Todorov et al., 2019), also in comparison with studies on clinical isolates (De Sousa, 2003). 12, 14 and 12 from 50 tested virulence genes were detected in *P. acidilacticii* strains ST1607V, ST2104V and ST3105V, respectively. In a previous study (Todorov et al., 2019) *P. acidilactici* ET35 isolated from smoked salmon was described to be positive for 14 virulence genes, including *cytA, efaA, ermA, ermB, ermC, tetO, aph(2")-Ic, vatE, bcrB, ccf, cob, int-Tn, vanA and vanC2*.

Studying virulence factors has always been a focus of investigation on pathogenic bacteria related to clinical isolates. Special attention has been given to *Enterococcus* spp. as some strains are considered as opportunistic pathogens. However, some other LAB, including *Pediococcus* spp., can carry virulence factors as well and even if they are not expressed, such strains have to be considered as potential health hazards since under specific conditions they may either express these virulence factors, or the genetic information can be transferred to other bacterial species. Moreover, the determination of virulence factors in LAB by molecular and phenotypic procedures needs to be considered as an important, compulsory aspect, due to the risk of genetic transfer, since these features are usually encoded by genes located in conjugative plasmids. LAB in general have a remarkable long history as beneficial organisms, used in different fermentation processes for centuries and have found increased application as probiotics in the last few decades (Martinez et al., 2012). However, some strains, especially belonging to species of th genera *Enterococcus* and *Streptococcus*, are considered as opportunistic pathogens and can be associated with some clinical cases (Goldstein et al., 2015).

Spreading of antibiotic resistance genes are strain and gene specific (Table 1). Resistant strains to different antibiotics have been also identified, and several genes encoding such resistance have been identified and studied; e.g., different erythromycin-resistance genes (*erm*) have been found in many species of LAB, as well as a number of tetracycline resistance genes – *tet*(K, M, O, Q, S, W) (Dicks et al., 2011). Rojo-Bezares *et al.* (2006) reported that *ant*(6), *aph* (3′)-IIIa and *tet*L genes were found in *Pediococcus* spp. strains. The aminoglycoside resistance gene *aac*(6')-Ie-*aph*(2'')-Ia has been reported for some *Pediococcus* species from animal and wine origin, including *P. pentosaceus* and *P. parvulus* (Tenorio et al., 2001). Genetic determinants for macrolide resistance [*erm*(AM) genes] have been reported for *P. acidilactici* strains (Tankovic et al., 1993), and one of these genes was found to be encoded on a 46-kbp non-transferable plasmid (Tankovic et al., 1993). An *erm*B gene has been associated with a plasmid in a *P. acidilactici* strain (Danielsen et al., 2007). Bacterial cells can use a variety of mechanisms to share and spread resistance determinants. In natural environmental conditions the main mechanisms for horizontal gene transfer in bacteria are conjugation and transduction via bacteriophages (Kleinschmidt et al., 1993). Even with only limited information available regarding the pediococci and native conjugation systems, this scenario is not impossible.

Formerly, the transfer of genetic material - plasmid and transposoms from LAB to LAB and from LAB to other Gram-positive or Gram-negative bacteria - has been reported (Dicks et al., 2011). These strains belonging to the genus *Enterococcus* are well known for their ability as natural acceptors during th conjugation process (Clewell and Weaver, 1989). This scenario may also be possible for *Pediococcus* strains as well.

Table 1. Presence/absence for virulence factors, antibiotic resistance and biogenic amine production in *Pediococcus acidilactici* strains ST1607V, ST2104V isolated from artisanal cheeses in Brazil.

	ST1607V	ST2104V	ST3105V
esp	+	+	-
efaA	+	-	+
ermB	-	+	-
ermC	-	-	+
tetL	-	+	-
tetO	-	+	-
ant(4')-Ia	+	+	+
vatE	-	+	-
bcrB	+	-	-
aac(6')-Ii	-	+	-
ccf	+	+	+
fsrA	+	-	+
fsrB	-	-	+
fsrC	-	+	-
vanA	-	-	+
vanC1	+	-	+
vanC2	+	+	+
vanC1(2)	+	-	+
vanC1/C2	+	-	-
odc	-	+	-
bcrR	+	-	+
hyl	+	+	-
mur2ed	-	+	-
mur2ef	-	+	-
sprE	-	-	+
asa1, aac(6')-Ie-aph(2")-Ia, aph(2")-Ic, aph(2")-Id, hdc1, hdc2, tdc, ddlE, bcrD, catA, ant(6)-Ia, aph(2")-Ib, aph(3')-IIIa, ace, cob, cpd, cyt, ermA, gelE, tetK, tetM, tetS, vanB, int	-	-	-

Positive results (+) for genes for virulence, biogenic amines and antibiotic resistance in *P. acidilacticii* strains ST1607V, ST2104V and ST3105V

4. CONCLUSION
P. acidilacticii strains ST1607V, ST2104V and ST3105V have been isolated from artisanal cheeses in Brazil, and showed bacteriocinogenic activity against *Listeria* strains. Attention needs to be given to safety aspects for each strain before recommendation as starter, bio-preservative or probiotic culture for human or animal application. Low level presence of virulence factors related genes can be considered as a positive feature for the studied LAB strains. However, additional *in vitro* and *in vivo* experiments need to be designed in a systematic approach for simulating food processing and/or GIT conditions for determining possible expression of the few virulence genes present.

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Oral Session | Food Safety
[4-1015-C] Food Safety (1)
Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room C (3rd room)

[4-1015-C-04] Sensitivity Comparison of Standard and real-time PCR Assay for Detection *Salmonella* Typhimurium and Enteritidis in Indonesian Chicken Carcasses

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Keywords: chicken carcasses, real-time PCR, Salmonella Enteritidis, Salmonella Typhimurium, sensitivity

Salmonella is a pathogenic bacterium that can cause serious illness to humans. Chicken carcasses have been reported contaminated by *Salmonella,* especially *S.* Typhimurium and *S.* Enteritidis. Rapid and sensitive method for detection and differentiation of both species is required. The objective of this study was to compare the sensitivity of standard and real-time PCR (rt-PCR) assay for detection and differentiation two *Salmonella* serovars using pre-enrichment and enrichment treatment. Primer from invA gene, specific Typhimurium protein (STM) and specific Enteritidis virulence plasmid (Prot6E) sequences were used for marker of *Salmonella* genus, *S.* Typhimurium and *S.* Enteritidis respectively. Sensitivity determination was carried out by artificially contaminated of each *Salmonella* serovars into chicken carcasses approximately amount of 10^5 CFU/mL. After contaminated, DNA was extracted by chelex100 method and subsequently one-tenth dilution up to lowest concentration. Sensitivity was determined based on the lowest amount of DNA remain detected then converted to the cells number. The result showed that rt-PCR was more sensitive for genus detection. Without pre-enrichment step, the limit detection of rt-PCR for *Salmonella* genus, *S.* Typhimurium and *S.* Enteritidis were equivalent with 3.8×10^1 cells, 4.1×10^3 cells, and 2.6×10^4 cells respectively, meanwhile the standard PCR for each were equivalent with 5.3×10^4 cells, 4.0×10^4 cells, 4.1×10^3 cells respectively. Both of methods were applied to detect 20 *Salmonella*-contaminated chicken carcasses; rt-PCR assay can detected all of positive samples, whereas standard PCR showed 3 false negatives results. However, both of method showed similar results for detection serovars *S.* Typhimurium and *S.* Enteritidis. Pre-enrichment step in buffer phosphate for 8 hours increased the sensitivity of standard PCR until less than 10 cells even for two serovars detection. It's concluded that analysis by standard PCR required pre-enrichment step for obtaining the sensitive result.

Sensitivity Comparison of Standard and real-time PCR Assay for Detection *Salmonella* Typhimurium and Enteritidis in Indonesian Chicken Carcasses

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Abstract

Salmonella is a pathogenic bacterium that can cause serious illness to humans. Chicken carcasses have been reported contaminated by *Salmonella,* especially *S.* Typhimurium and *S.* Enteritidis. Rapid and sensitive method for detection and differentiation of both species is required. The objective of this study was to compare the sensitivity of standard and real-time PCR (rt-PCR) assay for detection and differentiation two *Salmonella* serovars using pre-enrichment and enrichment treatment. Primer from invA gene, specific Typhimurium protein (STM) and specific Enteritidis virulence plasmid (Prot6E) sequences were used for marker of *Salmonella* genus, *S.* Typhimurium and *S.* Enteritidis respectively. Sensitivity determination was carried out by artificially contaminated of each *Salmonella* serovars into chicken carcasses approximately amount of 10^5 CFU/mL. After contaminated, DNA was extracted by chelex100 method and subsequently one-tenth dilution up to lowest concentration. Sensitivity was determined based on the lowest amount of DNA remain detected then converted to the cells number. The result showed that rt-PCR was more sensitive for genus detection. Without pre-enrichment step, the limit detection of rt-PCR for *Salmonella* genus, *S.* Typhimurium and *S.* Enteritidis were equivalent with 3.8×10^1 cells, 4.1×10^3 cells, and 2.6×10^4 cells respectively, meanwhile the standard PCR for each were equivalent with 5.3×10^4 cells, 4.0×10^4 cells, 4.1×10^3 cells respectively. Both of methods were applied to detect 20 *Salmonella*-contaminated chicken carcasses; rt-PCR assay can detected all of positive samples, whereas standard PCR showed 3 false negatives results. However, both of method showed similar results for detection serovars *S.* Typhimurium and *S.* Enteritidis. Pre-enrichment step in buffer phosphate for 8 hours increased the sensitivity of standard PCR until less than 10 cells even for two serovars detection. It's concluded that analysis by standard PCR required pre-enrichment step for obtaining the sensitive result.

Keywords: chicken carcasses, real-time PCR, *Salmonella* Enteritidis, *Salmonella* Typhimurium, sensitivity

11:15 AM - 11:30 AM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room C)

[4-1015-C-05] Development of Calculation Framework for Stochastic Prediction of Uncertainty and Variability in Survival Spore Numbers during Non-isothermal Inactivation by Second-order Monte Carlo Simulation

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Keywords: Predictive microbiology, Dynamic model, Stochastic model, Bootstrap method, Markov chain Monte Carlo, Weibull model, Thermal inactivation

Although thermal inactivation is one of the control measures for microbial contamination in foods, thermal processing at a high temperature or long-time heating induces chemical and physical reactions in foods that reduce product quality. To determine minimum processing condition, mathematical models need to appropriately describe the bacterial behavior during inactivation; the log-linear model, generally used, based on *D*-value (Decimal reduction time) would lead to overestimation or underestimation of thermal death time. In addition, the conventional kinetic predictive models disregard the variability and uncertainty of bacterial death behavior, although various mathematical models (log-linear or non-log-linear models) have been used to describe reduction behaviors during bacterial inactivation. In this context, "uncertainty" represents the lack of perfect knowledge of the parameter value; "variability" represents the true heterogeneity of the population that is a consequence of the physiological system. The importance of uncertainty and variability of biological and natural phenomena is widely recognized in the context of the guidance of quantitative microbial risk assessment. Few studies have been performed to separately evaluate and describe the variability and the uncertainty in population dynamics of microbial inactivation. This study aimed to develop a stochastic bacterial survival model considering both the "variability" and "uncertainty" from kinetic data. The developed model estimates the variance of survival behavior during non-isothermal inactivation. Furthermore, the estimated bacterial death behaviors were validated with the observed survival counts data. The spores of *Bacillus simplex* were heated under fifteen isothermal conditions (pH: 5.4, 5.8, 6.2, 6.6 and 7.0; Temperature: 80, 85 and 90° C); the changes in the survival spore count were experimentally determined. The uncertainty in fitted Weibullian parameters was estimated from statistical resampling one thousand replicates of the survival spore counts in each thermal condition by the non-parametric bootstrap method. One thousand replicates of the secondary models were described by estimated parameters. The secondary models describe the relationship between Weibullian parameters and pH and/or heating temperature. In contrast, individual cell heterogeneity of the death spore counts in an infinitesimal time interval was described as "variability" by Markov chain Monte Carlo (MCMC) method based on a binomial distribution. The second-order Monte Carlo (2DMC) simulation estimated the changes in the survival spore behavior during non-isothermal heating with both the variations. In parallel, the variance in survival kinetics of survival spore counts during non-isothermal heating was observed to compare with the prediction of survival counts. The uncertainty of the Weibullian parameter estimations and individual heterogeneity of the death spore counts in an infinitesimal time interval was successfully described. Furthermore, in all of three non-isothermal histories, the variances in the survival spore counts during processing periods were successfully described by the developed 2DMC model. The MCMC successfully described stochastic results in the bacterial reduction behaviors during not only isothermal but also non-isothermal from kinetic data. In conclusion, the 2DMC model enabling to describe both the uncertainty and variability in thermal inactivation will play an important role in appropriate risk assessment for bacterial survival.

Oral Session | Food Safety
[4-1015-C] Food Safety (1)
Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room C (3rd room)

[4-1015-C-06] Evaluation Growth Characteristics of Bacterial Spores Combine Treatment with High Hydrostatic Pressure and Alkaline Electrolyzed Water

*Koki Morita[1], Taiga Kuhara[1], Yoshinori Kamitani[1], Daisuke Hamanaka[1]　（1. Kagoshima University faculty of agriculture(Japan)）
Keywords: Bacterial spores, high hydrostatic pressure, Alkaline electrolyzed water, Injured microbes, Gonpertz curves, Antibiotics

In our previous study, we observed that the heat resistance of bacterial spores was effectively reduced by the combining treatment with high hydrostatic pressure(HHP) and alkaline electrolyzed water(AlEW). However, it is not clear in detail the effect of bacterial spores treated with AlEW and HHP, especially behavior of injured microbes which can live but cannot growth normally with the delay of lag phase etc. Since the existence of injured microbes may cause underestimate of viable counts comparing with the actual number of survived cell, correct understanding of growth characteristic is important to evaluate the microbiological safety of food. The growth curve of the microorganism is sigmoid and is divided into four phases; lag phase, accelerated log phase, slowed log phase and stationary phase. Gompertz curves are often used as continuous equation to represent sigmoidal growth curves. Growth of injured microbes is well known to affect growth characteristics, including delay of lag phase and reduction of growth rate as compared to those of intact one. In this study, we investigated the evaluation of injury characteristics by the double culture method supplemented with antibiotics in growth media, and growth behavior of bacterial spores treated by AlEW, HHP and their combination using Gompertz equation to obtain parameters in growth curve.Bacterial spores used in this study were *Bacillus subtilis* NBRC3134. Bacterial spores ware formed by conventional method and suspended in sterile sterile physiological saline (PS) to treat HHP.Contacting treatment of bacterial spores with AlEW was performed by suspending in test solution in plastic tube, and centrifuged. The pellet of bacterial spores on the bottom of plastic tube were re-suspended by vigorous pipetting and vortex-shaking. This process was performed two times, and treated spores were finally suspended with PS solution. PS solution was used as a control.Spore suspension was poured into plastic bag without air for following treatment.HHP treatments were performed with 100MPa for 1 hour at 50℃.The HHP treated pouch was aseptically opened, an appropriate amount of spore suspension was inoculated trypticase soy broth (TSB) supplemented with Chloramphenicol (CP) or Penicillin (PCG) for evaluation of the injury characteristics related to synthesizes of protein(enzyme) or peptidoglycan, and incubated at 30℃ for 48 hours. The optical density of the inoculated TSB at 490nm was measured every 30 minutes.Gompertz equation was fitted to the obtained OD data by nonlinear least squares method to obtain various parameters.At the same time, the growth behavior of AlEW and HHP treated bacterial spores were evaluated by calculating the length of lag phase and their difference between control and treated sample.So far, the investigation of the changes in growth characteristics of microorganisms have been reported and, it is known that shortening and extension of lag phase, fluctuation of growth rate in log phase, and difference in maximum viable count in stationary phase are affected. In this study, normal culture media and media supplemented with antibiotics were used as recovery media for injured cells produced by high-pressure-based treatments with or without AlEW, and their growth characteristics were compared with those of intact bacteria. The period of lag phase bacterial spores was not significantly different between high-pressure-treated and untreated sample, but significant extension of lag phase was obtained by the combination with AlEW. In addition, further extensions of lag

phase in growth curve were observed when a medium supplemented with CP or PCG was used. On the other hand, there were small differences between the treated and untreated samples in both the growth rate in log phase and maximum number of bacterial cell even in the case of CP or PCG supplementation. In general, extension of the lag phase in growth behavior is considered to be a process of recovery of injured bacteria. Moreover, the difference in growth rate in the log phase and the difference in the maximum number of viable cells in the stationary phase are considered to be the result of changes in the nutrient requirement of the microorganism related to the stress treatment. CP and PCG are antibiotics with features that inhibit protein and peptidoglycan synthesis, respectively. Considering the results obtained in this study, bacterial spores treated with high pressure treatment with and without AIEW require the time duration for recovering from damage associated with the synthesis of some essential biopolymers, but nutritional requirements and other phenotypic properties might not be affected.

Journal of Food Science and Agricultural Technology

Original Research Article

Evaluation Injury Characteristics of *Bacillus* Spores by Combination of Hydrostatic Pressure with Alkaline Electrolyzed Water

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ABSTRACT

The effect of combing procedures of high hydrostatic pressure (HHP) with alkaline electrolyzed water (AlEW) on the injury characteristics of bacterial spores were investigated. Effective reductions of survival *Bacillus subtilis* spores combined treatment of AlEW with HHP were obtained by the following heat shock at 80°C for 15 min. No survivals were observed by 100MPa of HHP treatment. Approximately 90 to 99% of bacterial spores treated by HHP with AlEW were injured or lost heat resistance since 1 to 2 logs difference in the survival comparing with the unheated bacterial spores were obtained. Double culture method (DCM) for assessing the injury characteristics of bacterial spores was performed by using trypticase soy broth supplemented with chloramphenicol (CP) and rifampicin (RFP), which are related to the synthesis inhibition of protein and RNA, respectively. Severe injury of the treated bacterial spores assessed by RFP supplemented broth were obtained at 50 and 100MPa with and without AlEW. Especially at 50MPa, considerable synergistic effect of AlEW on spore injury was observed. These results suggested that injury associated with the biosynthesis of some enzymes was observed at relatively low pressure level.

INTRODUCTION

Thermal processing is a major method for reducing the number of microorganisms in various food commodities. However, huge amount of thermal energy for inactivating bacterial spores, which are often existing in various agricultural commodities and has high resistance against physical stresses, results in the degradation of internal/external quality of fresh/processed foods such flavor, texture, surface color, functional components, and so on (Miyamoto et al., 2009). In order to reduce the negative effect on the quality of foods by thermal energy, various non-thermal antimicrobial technologies have been investigated for the actual application as an alternative. Whereas there are disadvantages about the inactivation process of bacterial spores (Ogino et al., 2015), high hydrostatic pressure (HHP) treatment has been focusing as the useful technology with small degradation of food

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quality.

Bacterial spores have high resistance against various chemical and physical stresses as described, it is well known that the injured or germinated bacterial spores induced by chemical/physical treatment lost their resistance against the following antimicrobial processes without severe condition (Sonoike et al., 1997). Several researchers reported that HHP also has a potential for reducing heat resistance of bacterial spores (Tsutido et al., 1992). Comparing with single heat treatment, additional 3-4 logs reduction of bacterial spores, previously treated by a 200MPa of HHP at 50°C for 10 min, was obtained by 80°C (Bartlett et al., 2002). The further reduction of pressure level is really important for considering about the application of HHP antimicrobial processing to the food industry by appropriate combining procedure of additional technologies.

In our previous study, we obtained the combination of HHP with alkaline electrolyzed water (AlEW), generated by electrolysis of diluted saline solution in a cathode side of diaphragm tank, has a potential to reduce the heat resistance of bacterial spores. Whereas the reduction of heat resistance was considered a result from spore injury by the combining treatment, the detailed mechanisms of the injury process had not been elucidated.

Double culture method (DCM) using standard culture media, such as trypticase soy agar; TSA, with/without typical growth-inhibiting agents has been used the determination of the number of the injured cell treated by an inactivation process. DCM is useful for the evaluation of the number of injured cell, however, there are some disadvantages for the clarification of detail mechanisms of injury process. For the further understanding of the injury characteristics of bacterial spores treated by AlEW with HHP, the modified DCM was required to be applied.

In this study, we investigated the evaluation of injury characteristic of bacterial spores treated by AlEW, HHP and their combination using DCM applied with antibiotics supplemented growth media.

MATERIALS AND METHODS

Test bacterium and preparation of spore suspension

Bacterial spores used in this study were *Bacillus subtilis* NBRC3134 obtained from National Institute of Technology and Evaluation (NITE). Stocked culture of *B. subtilis* were transferred to trypticase soy broth (TSB, Becton Dickinson and Company, USA), and cultured at 30°C for 24 hours. A 0.1 mL of cultured broth was applied and spread onto standard method agar (SMA, Nissui Pharmaceutical Co. Ltd., Tokyo) plates supplemented with 1g/L of $MgSO_4 \cdot 7H_2O$ and incubated at 30°C for 7 days for spore formation. Spores were collected by flooding the cultured agar surface with sterile physiological saline (PS) solution, and then scraping the surface with a sterile glass rod. Harvested spores were washed three times in sterile PS solution by centrifugation at 3500×g for 10min, followed by heat treatment at 70°C for 1 hour in order to kill the vegetative type cells. The spore concentration of prepared suspension was approximately 10^8-10^9 colony forming unit per milliliter (CFU/mL).
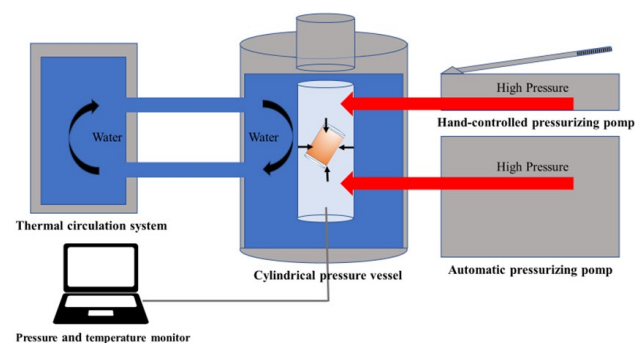
AlEW contracting treatment

Fig.1. Schematic diagram of pressurization apparatus

AlEW was generated using an electrolysis apparatus (ROX-20TA, HOSHIZAKI Electric Co. Ltd., Aichi, Japan). The current passing through the electrolysis apparatus was set at 10A, and the voltage between the electrodes was set at 10V. Oxidation reduction potential and pH were -835mV and 11, respectively. Physiological saline (PS) solution was used as a control water.

Contacting treatment of bacterial spores with AlEW was performed by suspending in test solution in plastic tube for 1 min, and centrifuged at 3500×g for 5 min. The pellet of bacterial spores on the bottom of plastic tube were re-suspended by vigorous pipetting and vortex-shaking for 3 min. This process was performed two times, and treated spores were finally suspended with PS solution.

HHP treatment

A 5 mL of spore suspension was poured into plastic bag without air, and aseptically heat sealed for preparing 2x5 cm^2 pouch for following treatment. HHP treatment was immediately performed by the pressurization apparatus (Fig. 1, Echigo Seika Inc., Niigata, Japan) with a cylindrical pressure vessel (inside volume = 300mL). Internal temperature of vessel was controlled by thermal circulation system (LBX-350, AS ONE, Co., Osaka, Japan) at 50°C. HHP conditions were prepared by the combined pressurization system equipped both automatic and hand-controlled pressurizing pump connected to the vessel. HHP treatments were performed with 30, 50 and 100MPa for 1 hour. Internal pressure and temperature during treatment were monitored by sensors equipped in the vessel. Rates of pressurization and depressurization were approximately 20MPa/sec and 100MPa/sec, respectively.

Evaluation of spore survival

The treated pouch was cooled down to ambient temperature immediately, and aseptically opened by sterile scissors. Treated suspension was transferred to glass test tube, and performed heat shock at 80°C for 15min in the electric heating block in order to kill the injured cell. A 1 mL of spore suspension was serially diluted by PS, and a 0.1mL was applied and evenly spread onto TSA plate. TSA plates were incubated at 30°C for 24 hours, and the survival counts were calculated by the number of colonies appeared after incubation.

Evaluation of injury characteristics of bacterial spores

The treated pouch was cooled down to ambient temperature immediately, and aseptically opened by sterile scissors. A 0.1 mL of spore suspension was inoculated TSB supplemented with chloramphenicol (CP, Wako Pure Chemical Co., Saitama, Japan) or rifampicin (RFP, Wako Pure Chemical Co.), for evaluation of the injury characteristics related to synthesizes of protein (enzyme) or RNA, respectively. The concentration of CP and RFP in TSB were 100ppm, which is the maximum level without influence on the growth of *B. subtilis* spores used in this study. After incubation for 24 hours, a 200mL of cultured media was transferred into microtiter plate, and the optical density (OD) at 600nm was measured by microtiter plate reader (Elx800UV, Biotek Instruments, Inc., Vermont, USA).

Injury characteristics of bacterial spores were determined by injury ratio (I_R) calculated by following equation;

$$I_R = \frac{N_I - N_{AI}}{N - N_A}$$

Where I_R is the injury ratio to the intact spores. N, N_I, N_A and N_{AI} are ODs of cultured pure TSB inoculated with untreated bacterial spores, cultured TSB supplemented with antibiotics inoculated with the untreated bacterial spores, cultured pure TSB inoculating with HHP treated bacterial spores, and cultured TSB supplemented with antibiotics inoculated with HHP treated bacterial spores, respectively. In case the calculated value of IR is more than 1, it was considered that some specific properties of bacterial spores were influenced by single or synergistic effect of AlEW, HHP and its combination.

All experiments were performed triplicate and the means of spore survival and injury ratio were calculated.

RESULTS AND DISCUSSION

It is well known that microbicidal efficiency of HHP treatment accelerate as increase in the pressure level. In this study, HHP treatment with 100MPa had a potential to inactivate bacterial spores comparing with that with 30MPa as shown in Figure 2. In addition, 0.5-1.5 logs reduction of spore survival were obtained by the combination of AlEW regardless of the difference in the pressure level. Approximately 90-95% of bacterial spores were injured and lost heat resistance by single AlEW or HHP and its combination under 50MPa, because the heat shock treatment at 80°C for 15min inactivate 1-1.5 logs of bacterial spores at 30 and 50MPa. On the other hand, all bacterial spores pressurized at 100MPa both with and without AlEW
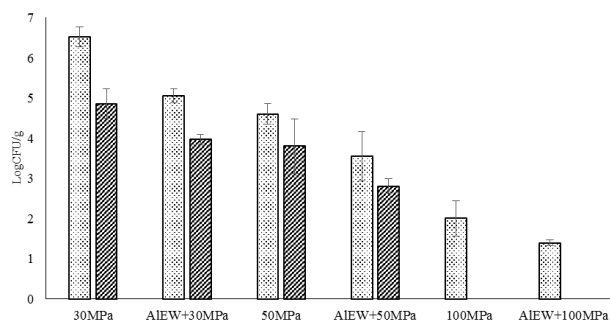

Fig. 2 Survival of bacterial spores treated with/without alkaline electrolyzed water and high hydrostatic pressure followed by heat treatment. ▨: before heating, ▨: after heating
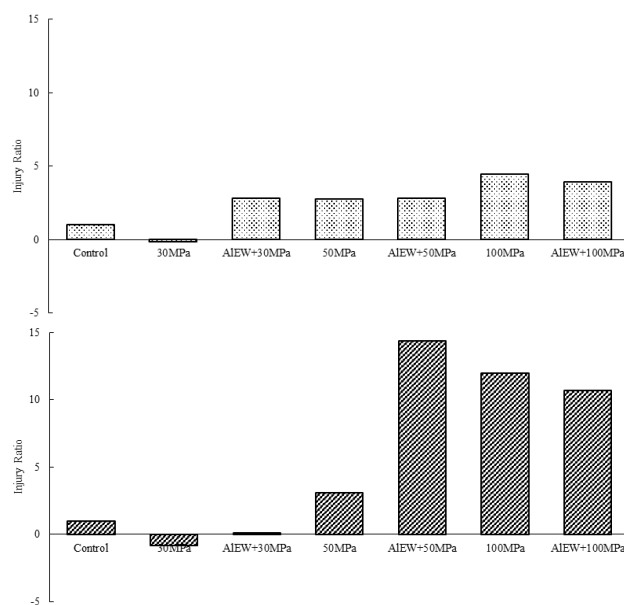

Fig. 3. Injury ratio of *Bacillus subtilis* spores treated by HHP with or without AlEW cultured with Chloramphenicol (▨) or Rifampicin (▨) supplemented trypticase soy broth.

could be injured and lost heat resistance since no survivals were obtained by a heat shock treatment at 80°C.

In DCM procedure, as the injury assessment of bacteria treated by some bactericidal technology, typical growth agar media salt-supplemented or water activity-controlled were generally applied for the recognition of the difference in the counted number of the appeared colonies. However, typical colony counting procedure was difficult to understand the injury characteristics of the stressed bacteria. Therefore in this study, the optical densities of growth culture broth supplemented with two antibiotics of CP and RFP before/after incubation were applied to the evaluation of injury characteristics of bacterial spores. CP and RFP have specific inhibiting influences on the protein (enzyme) and RNA synthesizes, respectively, and the injury characteristics could be assessed by the differences in the sensitivities of injured and intact bacterial cells.

Figure 3 shows the calculated injury ratio obtained from the measured OD values of TSB supplemented with CP and RFP. The higher sensitivities of treated bacterial spores against CP supplemented broth were obtained in comparison with the control sample, AlEW treatments did not have synergistic effect on the injury ratio at more than 50MPa. High injury ratios of the treated bacterial spores assessed by RFP supplemented broth were also obtained at 50 and 100MPa with and without AlEW. Especially at 50MPa, considerable synergistic effect of AlEW on spore injury was observed. These results suggested that injury related to the biosynthesis of proteins such as enzymes was observed at relatively low pressure level, whereas the injury related to RNA synthesis was influenced by slightly higher pressure level. Further studies are needed to clarify the mechanisms of the synergistic efficiencies observed at 30MPa with CP broth and at 50MPa with RFP broth. It was considered that AlEW synergistic effect could appeared under slightly stressed conditions for bacterial spores.

Although most of microorganisms are injured when exposed to

various stresses, it is well known that microbes have a potential to develop a specific function corresponding to the type of stress and recover as it can grow normally (Tsuchido et al., 1999). Considering about HHP treatment, typical injury of bacterial cells could be the enzyme denaturation and the destruction of cell membrane during physical process. The combination of AlEW might accelerate the injury of bacterial spores related HHP treatment.

CONCLUSIONS

In this study, it was obviously clarified that the HHP treatment is injured in synthesis of protein and RNA, and the combination of AlEW is increased the efficient of injury to bacterial spores. However, further investigations are strongly required for the clarification of the relationship between HHP and AlEW injury characteristics and reduction of heat resistance. The analysis of molecular level such as gene expression would be important to evaluate the detail mechanism of HHP injury. Introducing appropriate technology for the inactivation of bacterial spores using HHP has great potential for manufacturing various kinds of food with better quality and safety. The reduction of pressure level with ensuring food safety would reduce the cost, and expected to be applied to any food industries in near future.

ACKNOWLEDGEMENTS

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Oral Session | Food Safety
[4-1015-C] Food Safety (1)
Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room C (3rd room)

[4-1015-C-07] Impact of Mechanization Development on Women and Hired Labor Utilizations of Small-Scale Rice Farming Operations in Kampar Region, Indonesia

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Keywords: Mechanization development, women labor, hired labor, small-scale rice farming

Agricultural mechanization has been undergoing the development process through the replacement of human labor and draught animals with farm machinery particularly in rice farming system. This research examines the impact of mechanization development on women and hired labor utilization of small-scale rice farming operations in Kampar Region. Field surveys were conducted in two districts in Kampar Region, namely Kuok and Bangkinang during July-August 2018. Sixty rice women farmers were purposively selected for samples and they were interviewed personally to collect primary data. Data were analyzed using a descriptive–quantitative approach and simple regression techniques. As a result, the mechanization development in small-scale rice farming gradually reduced women and hired labor utilization and quickly occurred primarily on labor-intensive operations, such as land preparation, harvesting, threshing, and milling. Totally, the time requirement for performing rice farming operations was relatively high to account for 602.56 $hr.ha^{-1}$. Most of the hours were required to perform manually operations, including weeding (136.31 $hr.ha^{-1}$), transplanting (132.59 $hr.ha^{-1}$), and harvesting (99.77 $hr.ha^{-1}$). These operations required more time due to dominantly worked by women and hired labor with manual tools. On the other hand, the working hours used farm machines reduced significantly, accounting for 61.88 $hr.ha^{-1}$, 99.76 $hr.ha^{-1}$, 30.27 $hr.ha^{-1}$, and 45.92 $hr.ha^{-1}$ for ploughing, harvesting, threshing, and milling, respectively. The reduced working hours in rice farming created few off-farm activities in survey areas

Impact of Mechanization Development on Women and Hired Labor Utilizations of Small-Scale Rice Farming Operations in Kampar Region, Indonesia

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ABSTRACT

Agricultural mechanization has been undergoing the development process through the replacement of human labor and draught animals with farm machinery particularly in rice farming system. This research examines the impact of mechanization development on women and hired labor utilization of small-scale rice farming operations in Kampar Region. Field surveys were conducted in two districts in Kampar Region, namely Kuok and Bangkinang during July-August 2018. Sixty rice women farmers were purposively selected for samples and they were interviewed personally to collect primary data. Data were analyzed using a descriptive–quantitative approach and simple regression techniques. As a result, the mechanization development in small-scale rice farming gradually reduced women and hired labor utilization and quickly occurred primarily on labor-intensive operations, such as land preparation, harvesting, threshing, and milling. Totally, the time requirement for performing rice farming operations was relatively high to account for 602.56 $hr.ha^{-1}$. Most of the hours were required to perform manually operations, including weeding (136.31 $hr.ha^{-1}$), transplanting (132.59 $hr.ha^{-1}$), and harvesting (99.77 $hr.ha^{-1}$). These operations required more time due to dominantly worked by women and hired labor with manual tools. On the other hand, the working hours used farm machines reduced significantly, accounting for 61.88 $hr.ha^{-1}$, 99.76 $hr.ha^{-1}$, 30.27 $hr.ha^{-1}$, and 45.92 $hr.ha^{-1}$ for ploughing, harvesting, threshing, and milling, respectively. The reduced working hours in rice farming created few off-farm activities in survey areas.

Keywords: Mechanization development, women labor, hired labor, small-scale rice farming.

1. INTRODUCTION

Women have long been recognized as an important labor force in rural economies around the world. They hold a crucial role in agricultural and rural development (Petrics et al., 2018) and key players in the agricultural sector (Enete and Amusa, 2010; Kumar and Kumari, 2018). Women are involved in over half of the farm activities in most developing countries (Bayisenge et al., 2019) and encompass the largest percentage of the workforce in the agricultural sector (Assefa and de Roo, 2015). Women have made greatly contribution to about 43% of the agricultural labor force in the agricultural sector globally and it rises to be 70% in some developing countries (FAO, 2011: Team and Doss, 2011; Petrics et al., 2018). The figures ranged from 15- 81% in Southeast Asia and 42% in Indonesia (Paris, 2006). However, the current agricultural transformation toward mechanized farming system has had a different impact on women labor contribution in agricultural sector and changed rapidly in many parts of the world (Sisei, 2016).

The vital role of women in agricultural production of most developing countries particularly in rice farming operations has been reported by a number of studies (Enete and Amusa, 2010; Ajadi, et al., 2015; Baba et al., 2015; Cisco, 2016; Sisei, 2016; Amare and Endalew, 2016; Sims and Kienzle, 2016). They revealed that women have been involved in the whole farm operations from land preparation to post-harvest processing with different rates of contribution. However, the involvement of women in rice production varies from region to region and even within regions (Fonjong and Athanasia, 2007; Effiong et

al., 2015). Moreover, the involvement of women differs with difference of agro-production systems (Bala, 2010) and also varies from land preparation to post harvest operations (Sucharita and Bishnoi, 2018).

Traditionally, the roles of rural women were such as household maintenance, cooking, fetching drinking water, and fuel wood (Santhi et al., 2005), while in rice production included planting, weeding, harvesting, processing, and rice post-harvest activities (Waris, 2015; Sucharita and Bishnoi, 2018). For instance, in Western Uttar Pradesh, India, women have engaged in most of the agricultural operations except ploughing and their participation depend on the labor requirement for various crops (Baliyan, 2018). In Nigeria, women mostly participated in post farming operations such as harvesting, storage, watering, and transporting (Olawepo and Fatulu, 2012). In the same country, Yusuf et al. (2014) stated that the contribution of the women ranges from tasks as land clearing, land tilling, and planting, weeding, fertilizer/manure application to harvesting, food processing, and threshing.

The rapid development of agricultural mechanization today through the introduction of new technologies especially farm machinery has brought about changes in farming system in most developing countries. The important process of the mechanization development is to replace human labors and draught animals by mechanical devices (farm machinery) in farming activities (Simalenga, 2000; Chandrasekaran et al., 2008, Akinbamowo, 2013; Basu and Nandim 2014; Reddy et al., 2014; Paman et al., 2018). In Adamawa state, for example, the human power is gradually replaced by single axle multipurpose machines from land preparation to post- harvest (Mada and Mahai, 2013). According to Diao et al., (2014), the agricultural mechanization represents technology change through the adoption of mechanical power to undertake agricultural operations such as ploughing, harvesting, shelling, and planting. This development process may withdraw women labor early out of the farming activities because they commonly operate manual tools to perform farm works and eventually had a major impact on the demand and supply for farm labor (Schmitz and Moss, 2015).

Farm works physically demand much power and the working conditions are often harsh (Srivastava, 2006). Under these conditions, mechanization technology is essential for agricultural production and processing (Mlengera et al., 2015) in order to take the drudgery out of such hard work (Kienzle et al, 2013; Mujawamariya and Kalema, 2017; Sucharita and Bishnoi, 2018; Namdeo, 2018), which makes a difficulty for women labor to manually perform of the operations. Currently, the mechanization is required not only for increasing crop production, but also for processing and along the entire value chain (Sims and Kienzle, 2017). Furthermore and most importantly, mechanization can also help to perform for very difficult tasks in agriculture with short time and less cost (Basu and Nandi, 2014), so the introduction of mechanization implements and technologies enables to lighten burden of women (Amare and Endalew, 2016). Hence, farm machinery has become increasingly available on farm and highly required primarily for rice farming operations in rural area today.

The displacement of women labor and together with hired labor in agricultural activities as a result of mechanization development has been a common phenomenon in most developing countries, including Indonesia. The widespread use of the farm machinery has had serious equity consequences in terms of the displacement of labor and tenant farmers (Pingali, 2007). Kirui (2019) reported that tractor-powered mechanization had significant effects on the use of family and hired labor. In Indonesia, women still hold a central role in the agriculture activities in the country, particularly in rice production as well as post-harvest processing. The present research examines the impact of mechanization development on women and hired labor utilization in small-scale rice farming operations in Kampar Region.

2. MATERIALS AND METHODS

Kampar Region is divided into 21 districts that widely spread on area of 1,128,928 hectares. According to Food Crops Services of Kampar Region (2018), plantation area is dominant in the region to reach 415.702 hectares (approximately 37% of total), consisting of palm oil, rubber, coconut, crops, and other plantation crops, and the second dominant area is forestry to reach 76,853 hectares (6.81% of total). While, paddy field area has 6.546 hectares (0.58% of the total) and spreads only over 15 districts and the areas are mostly rain-fed paddy field. Although rice is not a main crop in the region, it is an important

crop to provide stable food for feeding population especially in the region. Therefore, local government greatly supports rice farmers to enhance production and productivity through more intensively application of farm machinery to perform farming operations.

Rice in Kampar Region is grown on both irrigated and rain-fed paddy field areas and cropping season can be once or twice a year depending on type of land and climatic conditions. Wet (rainy) season is main cropping season of rice in Kampar region i.e., during October – March. The second cropping season is on dry season, i.e., during April – August every year. The duration of both seasons included one month for land preparation before rice transplanting. Interview with farmers revealed that the whole rice areas are usually cultivated during wet season due to supply water from rainfall is sufficient mainly the beginning of growing season to facilitate land preparation.

From 21 districts in Kampar Region, there are fifteen districts to have paddy field areas that produce rice every year. These districts are important areas to supply local rice need in the region. The field survey was conducted only in the two districts, namely Kuok and Bangkinang which were purposively selected from fifteen districts of the region. Two villages then were selected from each selected districts with a total of four villages. The areas selected for the survey are based on their high level of farm machinery application with a high role of human and hired labor in the rice production activities.

The population of the research consisted of small rice farmers from four selected villages in the Kampar Region. Fifteen small rice farmers were randomly selected from each village for samples making a total of sixty rice farmers. Most of the samples are women who directly involve in rice farming cultivation. This research used primary data which were collected by personal interview method with the farmers using a structured questionnaire after the main cropping season of 2018. Data collected consisted of sample characteristics, working hours of men, women, hired labors, and machines for each type of rice farming operations, rice farming areas, and rice yields. The data collected thus were tabulated and analyzed by using descriptive-quantitative appropriate such as percentage, median and simple regression technique.

3. RESULTS AND DISCUSSION

3.1. Characteristics of samples and rice farms

As outlined in advance, sample farmers in this research were women who directly involved in rice farming cultivation. They were 46 years old and had 8 years of formal education on average. Rice cultivation has been carried out from generation to generation as the main source of livelihood and provided rice food mainly for household consumption. Therefore, the farmers have a long time of experience in rice cultivation to average 17 years. The number of family members was 5 persons on average, consisting of parent and three children. Most of farmers did not employ their children in rice farming activities because they go to school in their villages or in the city out of the village.

The rice farm owned farmers in survey areas is characterized by small-scale area, ranging from 0.11 to 1.00 hectares. Most farmers divided their farm areas into small plots with an average of 250 m^2 per plot (ranging from 107 m^2 to 2,500 m^2). This was done to be easier for doing tillage work when they previously used manual tool (hoe) and to make easily supply water into the field. Consequently, farmers slightly experienced a difficulty to perform tillage work by machine. Few farmers could not fully use the machine to perform the operations due to too small size of the farm holdings. The time requirement to complete the tillage work was still relatively longer as a result of the use of farm machines was still limited. Beside small farm size, the insufficient water supply into paddy field also constrained the application of farm machines for rice farming in the Kampar Region.

3.2. Labor and machine utilization

Type of farm machines which have popularly been used by small rice farmers in Kampar Region are hand tractors, combine harvesters, power threshers, and rice milling units (RMUs) (Figure 1). They have been gradually replacing traditional tools like hoes, sickles, pedal threshers, etc. with farm machines.

According to Paman et al. (2014), these farm machines are mostly managed by hire service providers for small rice farmers. There is not many farmers to own farm machines themselves due to low purchasing power. Therefore, majority of small farmers harnessed farm machinery service providers to work their farming operations. In practices, the application level of the machines in rice farming operations varied not only among districts or rice growing areas but also between individual farmers. The level of the machinery application among farmers depended on farmer's economic ability to pay machinery services, availability of family labors, size of farm holdings, and farmers' age. For instance, small farmers who have enough money replaced most family women and hired labor with farm machines. The main reason is to shorten operation time and improve work quality of rice farming. The using of farm machines enables farmers in the same area to plant rice at the same time to hinder pest attack like bird, rat etc. Consequently, the utilization of both women and hired labor gradually reduced in the survey areas.

Rice production is a labor-intensive operation with fluctuating workload depending on operation type of farming. In practices, labor is usually required much more and very hard for plowing, planting, harvesting, threshing, and milling operations. Therefore, these operations were replaced more early than other ones, like seedling, weeding, pest and disease control, transporting, and drying. In survey areas, these operations were worked by either human and hired labor or farm machines. It was found that approximately 90% of farmers fully used farm machines (hand tractor) to perform land preparation, whereas this operation was previously worked manually by either women or hired labor. About 20% and 72% of farmers used farm machines to perform both harvesting and threshing operations, respectively. The milling operation has fully mechanized and farmers did not employ women and hired labor for that operation anymore in the survey areas.

Figure 1. Type of farm machines used by small rice farmers di Kampar Region

Table 1 shows that totally, time requirement for rice farming operations was about 602 hr.ha^{-1}. This finding is smaller than 851 hr.ha^{-1} as reported by Paman et al. (2012) for Riau Province in 2012 (Kampar is one of 12 regions of the Riau Province). It means that there was a decrease of the required time for rice farming operations per hectare to approximately 29% during the last 7 years. Furthermore, the most time required for weeding was 136.31 hr.ha^{-1}, followed by transplanting (132.59 hr.ha^{-1}) and then harvesting (99.77 hr.ha^{-1}). These operations required more time due to dominantly worked by human labor. Hitherto, the farm machine for performing above operations have been not available yet, except for harvesting such as combine harvester.

Four labor intensive operations which have mostly been taken over by farm machines included plowing, harvesting, threshing, and milling. The operations were worked with requiring shorter time to account to 61.88 hr.ha^{-1}, 99.76 hr.ha^{-1}, 30.27 hr.ha^{-1}, and 45.92 hr.ha^{-1} for plowing, harvesting, threshing, and milling, respectively. Ploughing, harvesting, and threshing operations have not fully mechanized yet. The contribution of farm machines was about 61%, 3%, 67% for ploughing, harvesting, and threshing, respectively. Meanwhile, the milling was worked completely by small rice milling units (RMUs)/hullers. Now, both harvesting and threshing operations could be done together with combine harvester, so time requirement would be relatively shorter.

Table 1. Labor requirement for rice farming operations in hour per hectare

Type of operation	Family Labor		Hired Labor		Machine	Total
	Man	Woman	Man	Woman	(hr.ha^{-1})	(hr.ha^{-1})
Ploughing	13.74	14.80	0.00	0.00	33.34	61.88
Seedling	2.94	10.47	0.00	0.00	0.00	13.41
Transplanting	23.20	90.77	0.00	18.62	0.00	132.59
Fertilizing	3.30	34.41	0.00	0.00	0.00	37.71
Weeding	15.93	110.12	0.18	12.28	0.00	136.31
Pest and disease control	4.04	16.32	0.00	0.00	0.00	20.35
Harvesting	11.38	64.04	4.42	16.96	2.97	99.76
Threshing	0.00	9.87	0.00	0.00	20.40	30.27
Transporting	7.49	3.58	0.00	0.00	0.00	11.07
Drying	1.39	11.89	0.00	0.00	0.00	13.28
Milling	0.00	0.00	0.00	0.00	45.92	45.92
Total	83.41	356.40	4.60	47.86	82.23	602.56

Referring to Paman et al., (2015), there were three types of hand tractors have been popularly used for land preparation i.e., moldboard ploughs, rotary tillers, and hydro tillers. The use of these farm machines depend on field conditions primarily water supply on the paddy field. Both rotary tillers and hydro tillers were used under condition of the water supply into paddy field is sufficient where the water on the surface of field is flooding. Therefore, they were commonly used during wet growing season.

Furthermore, combine harvester has begun to be used for harvesting and threshing operations, although this machine is just in its infancy in survey areas today. Previously, the above operations were worked dominantly by women or hired labors with sickles for harvesting and pedal threshers for threshing. Consequently, the displacement of women and hired labors with machine caused to reduce significantly human labor utilization in rice farming operations. Interview with farmers revealed that land preparation required time at least 25 – 30 working days by human labor, this time reduced significantly to be only 2 - 3 days by hand tractors. Other operations that were also replaced by machines also required shorter and shorter time.
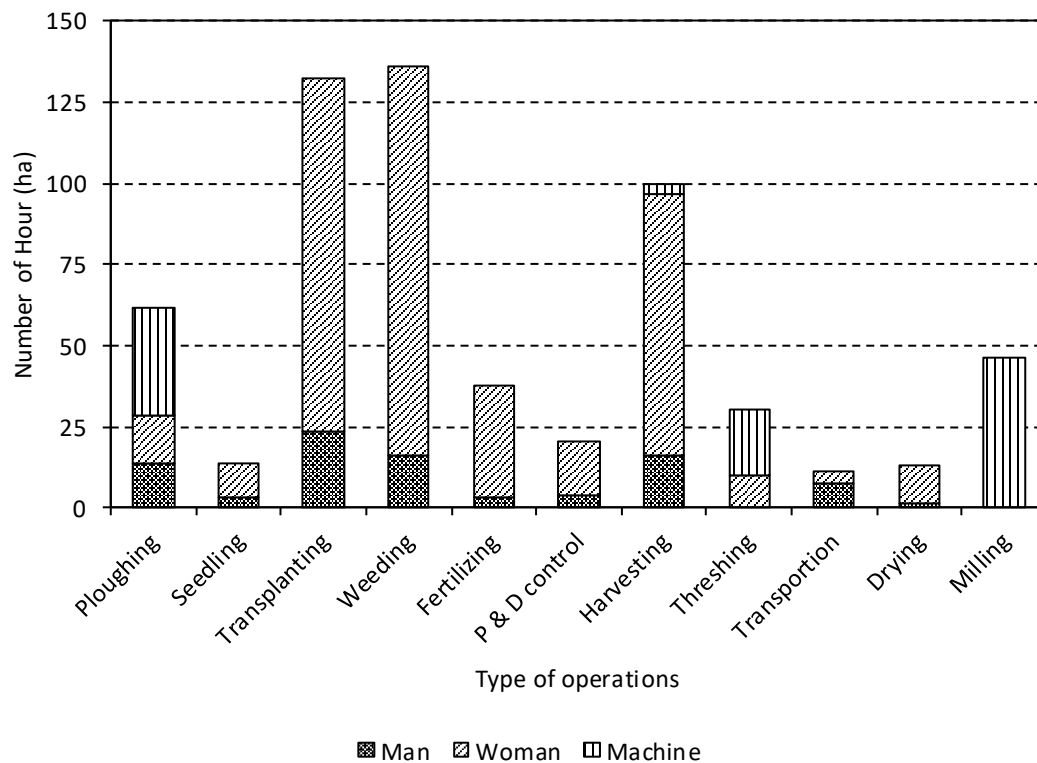
Figure 2. Contribution of human labor and machinery in rice farming operations

The contribution of woman labor as well as man labor in rice farming operations varied among types of operations as presented in Figure 1. The milling operation did not involve family women as well as hired labor anymore and the operation was completely worked by rice milling units (RMUs). This operation replaced by machine quicker than other ones and reached full mechanized today. It is because many milling service providers offering cheap price are available in survey areas. Plowing, harvesting, and threshing operations still involved women although their contributions have gradually being reduced. For instance, the contribution of women labor remained only 32% and 28% for plowing and threshing, respectively. Current introduction of mini tractors and combine harvesters will replace women labor soon in the future in survey areas. Other operations were still dominated by women, exception transportation.
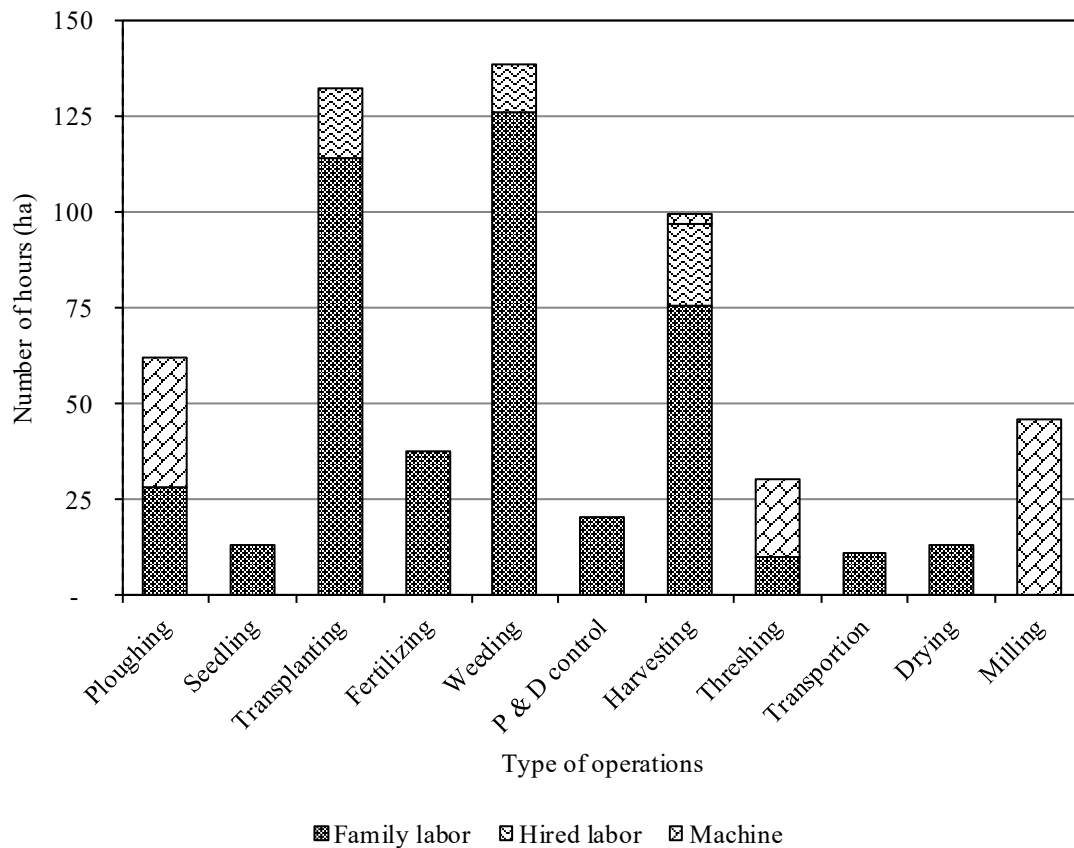
Figure 3. Contribution of hired labor and machinery in rice farming operations

According to Figure 2, the involvement of hire labors in rice farming operations in survey areas was only in transplanting, weeding and harvesting operations. Previously, the hired labors were mostly required in ploughing before being replaced by machine. While, for light and easy operations were commonly worked by family labors such seedling, fertilizing etc. The contribution of hired labor in transplanting, weeding and harvesting operations was relatively lesser to account for about 16%, 10%, and 28%, respectively. In this case, the hired labors were mostly woman, while men farmers were not so interested to be hired labor on rice farming due to low wage rate. The hired labors commonly worked 8 hours per day and were paid based on working day basis. The prevailing rates of wage which were established by farmer community in survey areas are IDR 100,000 (US $7) per working day for man labor and IDR 80,000 (US $6) per working day for woman labor under exchange rate of IDR 14,000 per US dollar. Most farmers revealed that the rate of wage did not increase during the last 4 years due to unchange significantly price of rice product.

The impact of mechanization development on both working hour of women and hired labors per hectare can be graphically illustrated in Fig. 4 and 5. The mechanization level in this analysis is the rate of machinery application in farming operations. A simple linear regression was used to relate between working hour of women and mechanization level. The linear regression could explain 45% ($r = 0.45$) of the observed variation in working hour of women and showed a negative relationship between working hour of women per hectare and mechanization

level as described by equation: $y = -0.030x + 24.401$. The result indicates that working hour of women per hectare tended to decrease with increasing mechanization level.
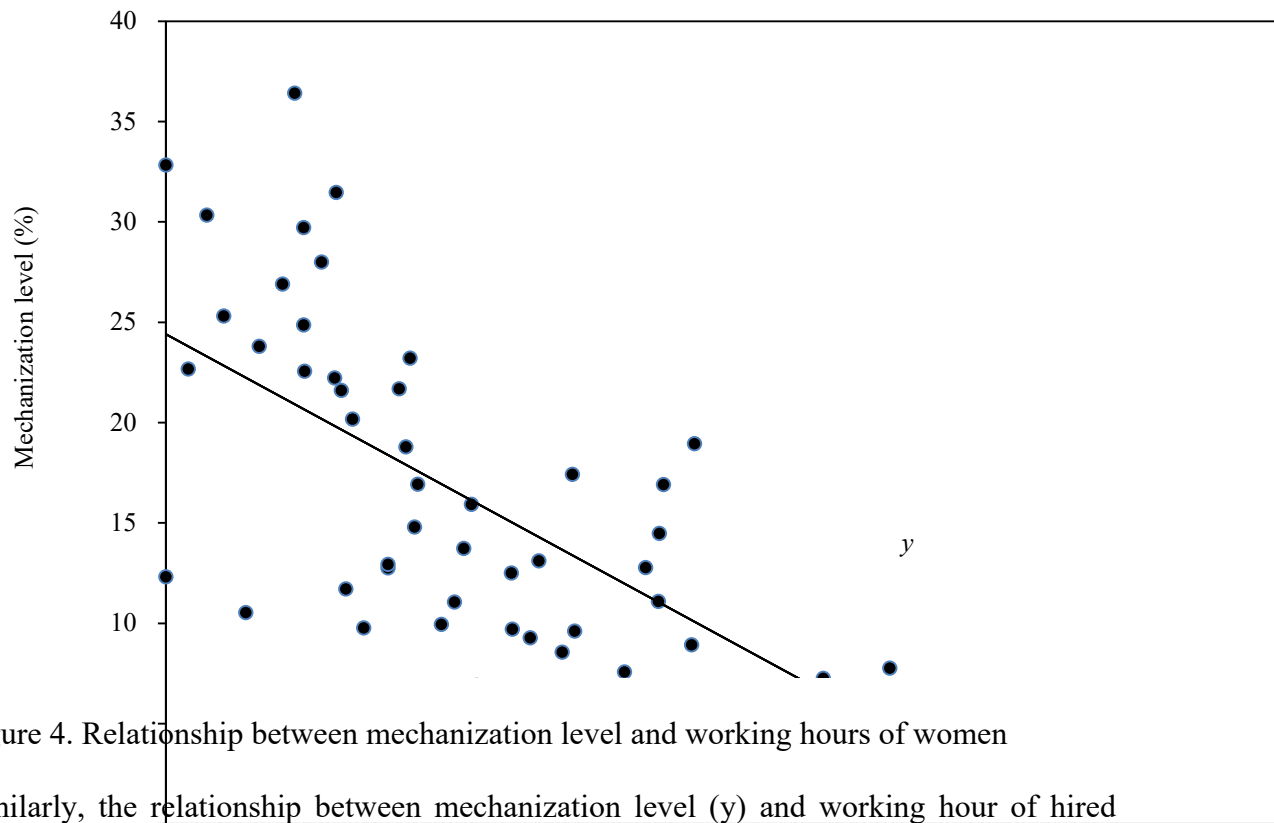

Figure 4. Relationship between mechanization level and working hours of women

Similarly, the relationship between mechanization level (y) and working hour of hired labors (x) per hectare also showed a negative and had a lower correlation. It means that the higher level of mechanization in farming operation caused the reduction of using hired labors. However, the decrease in working hour of hired labors could only explain 34% as shown by linear regression ($y = -0.038x + 20.119$; $r = 0.34$). The other factors that may cause the reduction of working hour of hired labors such as the rate of labor wage, availability of hired labors in the village, and etc. that did not include in the regression model.
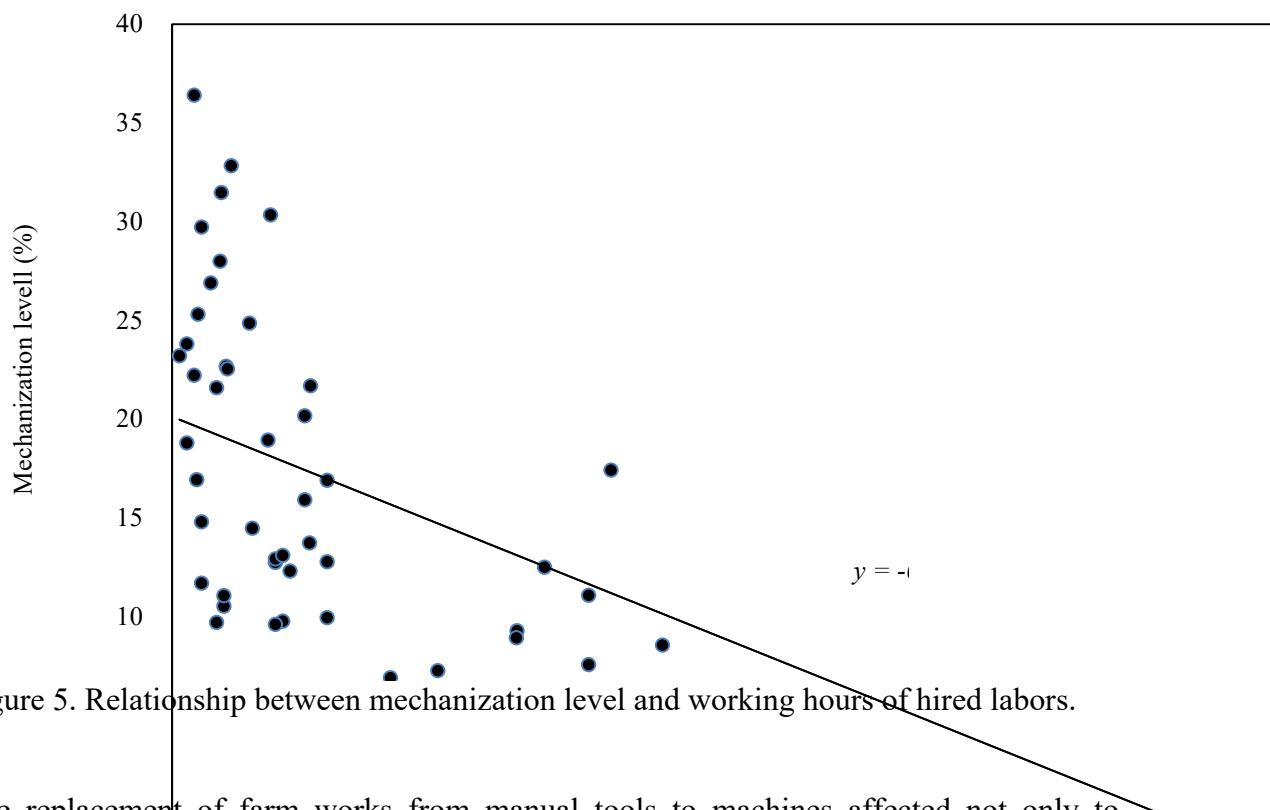
Figure 5. Relationship between mechanization level and working hours of hired labors.

The replacement of farm works from manual tools to machines affected not only to reduce time and work hard (drudgery) during rice growing but also to decrease farming costs and additional farmer's income for hired labor. The reducing cost of rice farming was significance and improved efficiency of farm management. Based on interviews with farmers revealed that cost of land preparation reached IDR 2,500,000 (US $143) per hectare by human labor. If this operation is worked by hand tractor, it was required only IDR 1,500,000 (US $107) per hectare or decreased about 25%. This cost referred to the rate of service charge for hiring machine from local farm machinery providers.

The reduced working hours in rice farming of women as well as hired labors lead them to create new job opportunities. They did off-farm activities such as fruit trading, making cake, etc. However, some women used the spare time to help their husband to work on non-rice farm activities such as oil palm, rubber, and orange garden. Furthermore, most of older women who aged more than 60 years old made use of time to take a rest. They felt tired to work in rice farming because this activity has been done since young (approximately 15 years old). They also wish to take the opportunity for participating much more in social and religion activities in their neighborhood.

4. CONCLUSIONS

The present research concludes that the rice farming operations have gradually used machines to replace women and hired labors primarily for labor-intensive operations, including land preparation, harvesting, threshing, and milling. The application rate of the farm machines was different between individual farmers as well as type of operations. Totally, the time requirement for performing rice farming operations was relatively high to be 602.56 hr.ha^{-1}. Most of the hours were required to perform manually operations such as weeding (136.31 hr.ha^{-1}),

transplanting (132.59 hr.ha^{-1}), and harvesting (99.77 hr.ha^{-1}). These operations required more time due to dominantly worked by women and hired labor with manual tools. On the other hand, the operation hours that used farm machines reduced significantly, accounting for 61.88 hr.ha^{-1}, 99.76 hr.ha^{-1}, 30.27 hr.ha^{-1}, and 45.92 hr.ha^{-1} for ploughing, harvesting, threshing, and milling, respectively. The working hours of women and hired labor per hectare tended to decrease with increasing farm machines application. Reduced working hours in rice farming has created few off-farm activities although some of them still work on non-rice farming and partly take a rest at home especially for older farmers.

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Oral Session | Food Quality

[4-1015-D] Food Quality (1)

Chair:Yutaka Kitamura(University of Tsukuba, Japan), Mizuki Tsuta(National Agriculture and Food Research Organization)

Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room D (4th room)

[4-1015-D-01] **Effects of Operational Conditions of Internal Combustion Furnace on rice husk Biochar and vinegar**

*WEI-PUO KUO[1], YUTAKA KITAMURA[2], Yoshiyuki HARA[4], CHING-CHEN HSIEH[3], YI-HUNG LIN[3], CHEN-PIN CHEN[1] （1. Taiwan Agricultural Machinery and Biomechatromics Engineering Technology Development Association(Taiwan), 2. University of Tsukuba(Japan), 3. National Pingtung University of Science and Technology(Taiwan), 4. Hokkaido Agricultural Experiment Station(Japan)）

10:15 AM - 10:30 AM

[4-1015-D-02] **Assessment of Red Tomato Freshness Using Ultraviolet-induced Fluorescence Image**

*Keiji Konagaya[1], Dimas Firmanda Al Riza[1], Minori Yoneda[1], Sen Nie[1], Takuya Hirata[2], Noriko Takahashi[2], Makoto Kuramoto[2], Tetsuhito Suzuki[1], Naoshi Kondo[1] （1. Kyoto Univ.(Japan), 2. Ehime Univ.(Japan)）

10:30 AM - 10:45 AM

[4-1015-D-03] **Thermal oxidation stability assessment of extra virgin olive oil using fluorescence and transmittance imaging system**

*Vincent Rotich[1], Dimas Firmanda Al Riza[1], Ferruccio Giametta[2], Tetsuhito Suzuki[1], Yuichi Ogawa[1], Naoshi Kondo[1] （1. Kyoto University(Japan), 2. University of Molise(Italy)）

10:45 AM - 11:00 AM

[4-1015-D-04] **Chalkiness Index of Sake Rice "Yamada Nishiki" Using Ultraviolet-Near-Infrared Transmission**

*Khokan Kumar Saha[1,2], Firmanda Al Riza Dimas[1], Yuichi Ogawa[1], Tetsuhito Suzuki[1], Naoshi Kondo[1], Takuma Sugimoto[3] （1. Lab of Bio-sensing Engineering,Graduate School of Agriculture, Kyoto University, Kitashirakawa-Oiwakecho,Sakyo-ku ,606-8502.(Japan), 2. Department of Agricultural Engineering, Bangabandhu Sheikh Mujubur Rahman Agricultural University,Salna,Gazipur-1706.(Bangladesh), 3. Senior Researcher, Hyogo Prefectural Agriculture, Forestry and Fisheries Technology Research Center, Addo City, Hyogo Prefecture ,671-3441.(Japan)）

11:00 AM - 11:15 AM

[4-1015-D-05] **Quantification of Tofu microstructure by image analysis**

*CHIANG WEN-HSIN[1], Yoshito SAITO[1], Kohei OGATA[1], Tetsuhito SUZUKI[1], Naoshi KONDO[1] （1. Graduate School of Agriculture, Kyoto University(Japan)）

11:15 AM - 11:30 AM

[4-1015-D-06] **Processing of Green Tea Paste by Micro Wet Milling and Quality Evaluation During Storage**

*Md Zohurul Islam[1], Yutaka Kitamura[1], Mito Kokawa[1], Shinya Fujii[2], Hisayuki Nakayama[2] （1. Graduate School of Life and Environmental Sciences, University of Tsukuba, Ibaraki, Tsukuba-shi, Japan(Japan), 2. Nagasaki Agricultural and Forestry Technical Development Center, Nagasaki, Japan(Japan)）

11:30 AM - 11:45 AM

[4-1015-D-07] Quality Changes During Ripening of Mango (*Mangifera indica* L. ' Nam Dok Mai') under Different Temperature Conditions

*Eriko Yasunaga[1], Kohei Nakano[2], Busarakorn Mahayothee[3], Pramote Khuwijitjaru[3], Shinji Fukuda[4], Wolfram Spreer[5] （1. The University of Tokyo(Japan), 2. Gifu University(Japan), 3. Silpakorn University(Thailand), 4. Tokyo University of Agriculture and Technology(Japan), 5. Hohenheim University(Germany)）

11:45 AM - 12:00 PM

10:15 AM - 10:30 AM　(Wed. Sep 4, 2019 10:15 AM - 12:00 PM　Room D)

[4-1015-D-01] Effects of Operational Conditions of Internal Combustion Furnace on rice husk Biochar and vinegar

*WEI-PUO KUO[1], YUTAKA KITAMURA[2], Yoshiyuki HARA[4], CHING-CHEN HSIEH[3], YI-HUNG LIN[3], CHEN-PIN CHEN[1]　(1. Taiwan Agricultural Machinery and Biomechatromics Engineering Technology Development Association(Taiwan), 2. University of Tsukuba(Japan), 3. National Pingtung University of Science and Technology(Taiwan), 4. Hokkaido Agricultural Experiment Station(Japan))
Keywords: Rice husk, ICF Internal Combustion Furnace, Bio vinegar , Biochar, Sustainable Agriculture

Traditionally, it is very difficult to make rice hulls useful industrial or agricultural products. Most of them are used as the bedding for cattle farms or even burned as fertilizers. The composition of rice hulls is actually very unique in nature. It contains approximately 20% opaline silica in combination with a large amount of the phenyl propanoid structural polymers called lignin. This abundant agricultural waste has all of the properties qualified with excellent insulating materials. Recent researches revealed that rice hulls are not very flammable and are highly resistant to moisture penetration and fungal decomposition. Moreover, rice hulls do not transfer heat very well, smell or emit gases, and are not corrosive with respect to aluminum, copper or steel. In the natural form, the rice hull is classified as a Class A or Class I insulation material, and therefore, can be used very economically to insulate the wall, floor and roof cavities of a house.
In order to extent the application of rice hulls, an innovative Internal Combustion Furnace (ICF) was developed to produce two type of bio-products, such as rice husk biochar and rice husk vinegar, respectively. Rice husk was produced by dried rice hulls that is firstly anaerobic smoked in ICB. Carbonized rice husks or biochar and rice husk vinegar are produced at the same time, which can be recycled in farmland. This approach made significant contributions to nature's sustainable management of agricultural wastes.
In Taiwan. However, the optimal conditions for operating ICF, including temperature, time, and composition of raw materials, are necessary to be determined, so that the good quality and quantity of biochar and biovinegar can be maintained. In this experiment, the ratio of gray matter and carbon was measured to understand carbonization rate as well as the quality of biochar and biovinegar with varying the opening of damper or the furnace air intake. In particular, 9 experiments on burning carbonized rice husks were conducted at 5, 7.5 and 10 mm opening of the damper. Along with the experimental chart, we could figure out that chimney temperature (front and end) and combustor temperature (top and bottom) have a positive relation to the opening width. The amount of biovinegar with cooling water is more than the one without cooling water. The wider the damper got, the higher the ratio of carbon and dust or ash volume.

10:30 AM - 10:45 AM　(Wed. Sep 4, 2019 10:15 AM - 12:00 PM　Room D)

[4-1015-D-02] Assessment of Red Tomato Freshness Using Ultraviolet-induced Fluorescence Image

*Keiji Konagaya[1], Dimas Firmanda Al Riza[1], Minori Yoneda[1], Sen Nie[1], Takuya Hirata[2], Noriko Takahashi[2], Makoto Kuramoto[2], Tetsuhito Suzuki[1], Naoshi Kondo[1]　(1. Kyoto Univ.(Japan), 2. Ehime Univ.(Japan))
Keywords: tomato (*Solanum lycopersicum*), storage, fluorescence image, color image, RGB values

A tomato (*Solanum lycopersicum*) is harvested with a yellow, pink or red color, although it turns red when it reaches to a consumer. The red tomato is better in taste. Thus, the agriculture near an urban area make use of the red tomato at the harvest in some countries including Japan. However, the color of red tomato does

not change. Thus, it is difficult to distinguish a degraded tomato from a fresh one. This is a limitation of a color for an indicator of tomato freshness. In contrast, ultraviolet (UV)-induced fluorescence provides another color information of tomatoes. In this study, we investigated a potential of tomato fluorescence to monitor the tomato freshness during the storage. Tomatoes (cultivar: Momotaro) were harvested on June 19th, 2018 at greenhouse in Ehime University with a red color. The total of 50 tomatoes were stored at 4 and 25° C for 8 d. A halogen lamp and UV light emitting diode (LED, 365 nm) were used for light sources of color and fluorescence images, respectively. A high-resolution CMOS camera EOS Kiss x7 (Canon Inc., Japan) with parameters set as ISO 100, F-6.3 and shutter exposure 1/25 s (for color images) and 4 s (for fluorescence image) was used. The color and fluorescence images were captured during the storage. The efficacies of these techniques were discussed in terms of the possible evaluation period and the sensitive color channel. In the color image, tomato color changed from a red to a deeper red, while in the fluorescence image the color changed from a blue to a blue-white gradually. At 4° C, the changes in both colors were relatively small compared with 25° C storage. To quantify the chromaticity, RGB values of each image was calculated and then expressed as its ratio (such as R/(R+G+B)) followed by the normalization using its initial mean value. In the color image, the R and G ratio changed rapidly within the initial 2 or 4 d, while in the fluorescence image, the G ratio changed up to 8 d. The reasons of these changes are also important for the application of this technique to the field. The origin of the color image was assigned to the lycopene synthesis, as shown in our extracts. In contrast, the excitation-emission matrix (EEM) of tomato pericarp suggested the origin of fluorescence images. EEM exhibited no fluorescence peak of lycopene in visible region. This was reasonable, since carotenoid is known to be week in the fluorescence. There also existed no peak of chlorophyll. This is also reasonable, since in the past studies, it is known that there are few chlorophylls in red tomatoes near the detection limit of high-performance liquid chromatography (HPLC). Hence, the possible origin of fluorescence image was some phenolics including flavonoid since phenolics are abundant in the tomato skin. Overall, the fluorescence image (the G ratio) was effective to monitor the tomato freshness for entire 8 d, while the color image (the R and G ratio) was effective only for the initial 2 or 4 d. As the most transportation and storage process happen in a cold chain with an ambient air, this study would help the development of monitoring system after the harvest before consumption.

10:45 AM - 11:00 AM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room D)

[4-1015-D-03] Thermal oxidation stability assessment of extra virgin olive oil using fluorescence and transmittance imaging system

*Vincent Rotich[1], Dimas Firmanda Al Riza[1], Ferruccio Giametta[2], Tetsuhito Suzuki[1], Yuichi Ogawa[1], Naoshi Kondo[1]　(1. Kyoto University(Japan), 2. University of Molise(Italy))
Keywords: Thermal stability, Extra virgin olive oil, Fluorescence, Transmittance, Imaging system

Extra virgin olive oil is a high-quality product with profound health benefits but is susceptible to degradation due to oxidation. Environmental conditions such as temperature, oxygen, and light promote the oxidation process. Thermal oxidation stability is of primary concern regarding food quality and safety. The ability to resist oxidation guarantees both nutritional and economic value. In this study, the thermal oxidation stability of four mono-cultivars of extra virgin olive oil from different regions of Italy was studied. The samples were placed under thermal treatment at 120° C and measurements done at equal time intervals for 180 minutes. To develop a simplified imaging system, the fluorescence characteristics of samples at the different duration of thermal exposure was measured using front-face fluorescence spectroscopy. An imaging system based on

fluorescence and transmittance was developed to assess the changes that occur due to thermal exposure. The quality indices including; Peroxide value, acidity, K232, and K270 were measured following IOC (International Olive Council) standard procedures. Image processing of both colour and fluorescence images was done to ascertain the cultivar response to the thermal treatment. The fluorescence peak regions for polyphenols, oxidation products, and chlorophyll were monitored, and a comparison made between the different cultivars. New fluorescence peaks were formed at emission wavelengths 435nm and 465nm, and 570nm, suspected to be products of oxidation and hydrolysis respectively. The cultivars with a high concentration of polyphenols showed greater resistance to the formation of oxidation products. The b* component of CIE L*a*b* colour space was identified to monitor the colour changes due to thermal exposure and to ascertain the thermal stability of different cultivars of extra virgin olive oil.

11:00 AM - 11:15 AM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room D)

[4-1015-D-04] Chalkiness Index of Sake Rice "Yamada Nishiki" Using Ultraviolet-Near-Infrared Transmission

*Khokan Kumar Saha[1,2], Firmanda Al Riza Dimas[1], Yuichi Ogawa[1], Tetsuhito Suzuki[1], Naoshi Kondo[1], Takuma Sugimoto[3] (1. Lab of Bio-sensing Engineering,Graduate School of Agriculture, Kyoto University, Kitashirakawa-Oiwakecho,Sakyo-ku ,606-8502.(Japan), 2. Department of Agricultural Engineering, Bangabandhu Sheikh Mujubur Rahman Agricultural University,Salna,Gazipur-1706.(Bangladesh), 3. Senior Researcher, Hyogo Prefectural Agriculture, Forestry and Fisheries Technology Research Center, Addo City, Hyogo Prefecture ,671-3441.(Japan))
Keywords: White core, Transmittance, Chalky thickness , Sake rice

White core refers to the opaque area in the kernel which is an important consideration for grading the raw material (sake rice) in the brewing industry. There is a relation between white core level and polishing ratio for sake rice to make premium quality sake. Currently, the evaluation of white core level in grains of sake rice is performed by the internal chalky thickness which is a destructive method. The aim of this research is the grading of sake rice chalkiness in non-destructively. Non-white core and white core [light, medium] and chalky rice, cultivar "Yamada Nishiki" , were used for this experiment. In this research, the transmission optical property in the ultraviolet-near-infrared (UV-NIR) region of rice was measured in order to characterize the suitable wavelength region in sake rice. The transmission optical property was measured by Jasco V-670 spectrophotometer equipped with an integrating sphere (ISH-723). The thickness of the chalky part was measured destructively. The evaluation of transmission optical property was done by image processing techniques. The images acquired by transmission mode of 4 types of intact sake rice kernel by using ring UV LEDs, Blue LEDs, Green LEDs, Red LEDs, and NIR LEDs in which the peak wavelength of the LEDs was 365nm, 465nm,525nm,630nm, and 830 nm respectively. The rice samples were more penetrated by light in the NIR region. The result indicates that there is more contrast between white core and non-white core rice in NIR image and followed by the red, green, blue and UV region images. Therefore, the wavelength region in NIR showed better discrimination between white core and non-white core sake rice. Finally, four images features; the ratio of the area (opaque area to full area), transmittance gray level intensity, the ratio of axis length in opaque (minor axis length to major axis length of opaque) and distance of centroid (opaque and full) were extracted from NIR transmittance images. The proposed chalkiness index (ratio of chalky thickness to whole kernel thickness) was inversely correlated with NIR transmittance image gray level intensity. The classification accuracy of the white core and chalky rice was 94.34% by support vector machine (SVM). Therefore, these results can be used to developed machine vision-based white core detection of sake rice.

11:15 AM - 11:30 AM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room D)

[4-1015-D-05] Quantification of Tofu microstructure by image analysis

*CHIANG WEN-HSIN[1], Yoshito SAITO[1], Kohei OGATA[1], Tetsuhito SUZUKI[1], Naoshi KONDO[1] （1. Graduate School of Agriculture, Kyoto University(Japan)）
Keywords: Tofu, microstructure quantification, image analysis, stiffness

Coagulation of soymilk, a complicated process in the production of tofu, is a critical step that complex factors, such as coagulant concentration, cooking temperature, and coagulant type, involved may affect the physical properties of tofu. The microstructure of bio-aggregates is fundamental to their physical properties (Lawrence *et al.,*2017). Until today, however, there are few researches on the relationship between tofu physical property and microstructure and only qualitative descriptions of tofu curd have been recorded. The aim of this study was to quantitatively and objectively evaluate tofu microstructure using image analysis and verify the relationship between tofu microstructure and stiffness while varying coagulant concentration. The tofu samples were made with varying coagulant concentration. These samples were then photographed using SEM for imaging analysis. In this research, Harilick textural features were calculated to quantify the microstructure of tofu curd and selected by Principal Component Analysis (PCA). 3 geometric parameters (number of holes per area, size of holes(area), and porosity) and selected Haralick textural features were finally correlated with Young's modulus to verify the relationship between tofu microstructure and stiffness. The proposed methods, Haralick texture analysis and microstructure quantification, have potential discrimination application in tofu SEM image. As coagulant concentration increased, the number of holes and sum average feature also increased, however porosity decreased. From these findings, it was observed that the number of holes, porosity and sum average feature are candidate features for tofu microstructure quantification of SEM images. Moreover, correlations between stiffness with porosity and sum average feature were negative and positive respectively. Although the two tendencies were observed, in the future, more samples with varying concentration are necessary to improve the results.

11:30 AM - 11:45 AM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room D)

[4-1015-D-06] Processing of Green Tea Paste by Micro Wet Milling and Quality Evaluation During Storage

*Md Zohurul Islam[1], Yutaka Kitamura[1], Mito Kokawa[1], Shinya Fujii[2], Hisayuki Nakayama[2] （1. Graduate School of Life and Environmental Sciences, University of Tsukuba, Ibaraki, Tsukuba-shi, Japan(Japan), 2. Nagasaki Agricultural and Forestry Technical Development Center, Nagasaki, Japan(Japan)）
Keywords: Green Tea Paste, MWM, Color, Antioxidants, Storage study, Kinetic model

Green tea is a delicious variety of tea that is made from the *Camellia sinensis* plant. It is a non-fermented tea, thus contain higher amounts of phytochemicals, alkaloids, polyphenols including EGCG compared with black tea and oolong tea. In this regard there is increasing interest in its health benefits has led to the inclusion of green tea in the processing of value-added products with functional properties. But there are challenges to add green teas into the process product due to the higher particle sizes and insolubility in cold water. Therefore, the aim of the present study was to develop green tea paste by micro wet milling system with minimum particle sizes which lead to increase solubility and enhance bioactive compounds. In the present

study, three varieties of green tea samples Yabukita (shaded) Yabukita (non-shaded) and Hoji cha were collected from Nagasaki, Japan. The green tea paste was produced by MWM. The wet milling conditions were set by varying the green tea to water ratio (10:90; 15:85; and 20:80 w/w), feeding rate (15 mL/min to 25 mL/min) and constant rotational speed of 50 rpm for the preparation of green tea paste. The optimum milling conditions were determined based on achieving minimum particle sizes. Feeding rate 20 mL/min, green tea to water ratio of 20:80 w/w, and rotational speed 50 rpm can able to produce paste with smaller average particle sizes of 58.64±2.31 µm with better color (in terms of greenness and chroma value) and nutrition properties (i.e. ascorbic acid, polyphenol, and antioxidants). To evaluate the shelf life and storage stability, three kinds of green tea paste were stored at 20 ° C, 4 ° C, -18 ° C and -60 ° C with vacuum packing for 4 weeks. Temperature and storage time negatively influenced the stability of ascorbic acid, color, and antioxidant activity during storage. A kinetic study of the green tea paste was conducted to quantify the losses occurring in ascorbic acid, antioxidant activity and changes in the color of the green tea paste. The study revealed that the logistic model can predict the variation in ascorbic acid and antioxidant activity with higher R^2 = 0.98 value. However, first-order kinetic models were found suitable to predict the changes occurring in ascorbic acid, antioxidant activity and color properties (L, a, b, chroma). Whereas the total color changes (Δ E) showed a good fit with zero order kinetic models (R^2 = 0.98). So we concluded that we can preserve the green tea paste at a lower temperature by keeping minimum losses of color and antioxidant properties. Before commercialization, a sensory and microbial study needs to be carried out in the future.

11:45 AM - 12:00 PM (Wed. Sep 4, 2019 10:15 AM - 12:00 PM Room D)

[4-1015-D-07] Quality Changes During Ripening of Mango (*Mangifera indica* L. ‘Nam Dok Mai’) under Different Temperature Conditions

*Eriko Yasunaga[1], Kohei Nakano[2], Busarakorn Mahayothee[3], Pramote Khuwijitjaru[3], Shinji Fukuda[4], Wolfram Spreer[5]　(1. The University of Tokyo(Japan), 2. Gifu University(Japan), 3. Silpakorn University(Thailand), 4. Tokyo University of Agriculture and Technology(Japan), 5. Hohenheim University(Germany))
Keywords: accumulated respiration, climacteric rise, chilling injury, sucrose contents, hardness

The purpose of this study was to clarify the influence of storage temperature on the relationship between the accumulated respiration and the quality change during the ripening of unripe (green) mango fruits (*Mangifera indica* L. ‘Nam Dok Mai’). Unripe mango fruits were placed in an incubator at 10 ° C and 25 ° C to measure respiration rate and quality changes for 6 days. Postharvest ripening of unripe mango fruits was observed as changes in fruit firmness, peel color, brix, acidity, citric acid content, and sugar content under the storage conditions. The respiration rate of mango fruits stored at 25 ° C gradually increased from day 2 with a peak of climacteric rise on day 5, while climacteric rise for fruits stored at 10 ° C was not observed. It was confirmed from the respiration rate that post-ripening was not promoted at 10 ° C. The hardness, peel color, sucrose content and citric acid content of the fruit stored at 25 ° C decreased rapidly after day 2, while there was almost no change in the fruit stored at 10 ° C for 6 days. The accumulated respiration the fruit stored at 25 ° C for two days was equivalent to the fruit stored at 10 ° C for 6 days, both showing the same value in all quality indicators measured in this study.

[4-1330-A] Postharvest/Food Technology and Process Engineering (2)

Chair:Olaniyi A. Fawole(Stellenbosch University, South Africa), nutthachai pongprasert(King Mongkut's University of Technology Thonburi, Thailand)
Wed. Sep 4, 2019 1:30 PM - 3:30 PM Hall A (Main Hall)

[4-1330-A-01] Edible Coatings Control Shrivel and Maintain Quality of Nectarines during Simulated Export Conditions

Shannon Riva[2], *Olaniyi Amos Fawole[1], Umezuruike Linus Opara[1,2] （1. Postharvest Technology Research Laboratory, South African Research Chair in Postharvest Technology, Department of Food Science, Stellenbosch University(South Africa), 2. Postharvest Technology Research Laboratory, South African Research Chair in Postharvest Technology, Department of Horticultural Science, Stellenbosch University(South Africa)）
1:30 PM - 1:45 PM

[4-1330-A-02] Development and Characterization of Chitosan Film Incorporated with Cashew (*Anacardium Occidentale*) Leaf Extracts

*moooksupang - Liangpanth[1,2,3] （1. Mae Fah Luang University(Thailand), 2. Thomas Karbowiak(France), 3. Wirongrong Tongdeesoontorn(Thailand)）
1:45 PM - 2:00 PM

[4-1330-A-03] Green Synthesis of Zinc Oxide Nanoparticles from Asiatic Pennywort (*Centella asiatica* L.) and Its Effect on the Rice Starch-Gelatin Composite Film

*Wantida Homthawornchoo[1,2], Suttiporn Pinijsuwan[1,2], Saroat Rawdkuen[1,2] （1. School of Agro-Industry, Mae Fah Luang University(Thailand), 2. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University,(Thailand)）
2:00 PM - 2:15 PM

[4-1330-A-04] Combination of high pressure processing and heat treatment on quality and antioxidant activity of fresh-cut persimmon

Paweena Jarungjitaree[1], Matchima Naradisorn1[1,2], Daisuke Hamanaka [3], *Sutthiwal Setha [1,2] （1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, 57100, Thailand(Thailand), 2. Research Group of Postharvest Technology, Mae Fah Luang University, Chiang Rai, 57100, Thailand(Thailand), 3. Faculty of Agriculture, Kagoshima University, Kagoshima, 8900065, Japan(Japan)）
2:15 PM - 2:30 PM

[4-1330-A-05] Postharvest Ethylene Application Influences Biochemical Quality of Pummelo Fruit Under Low Temperature Storage

*Paemika Promkaew[1], Varit Srilaong[2], Satoru Kondo[1] （1. Graduate School of Horticulture, Chiba University(Japan), 2. Division of Postharvest Technology, School of Bioresources and Technology, King Mongkut's University of Technology Thonburi (Bangkhuntien)(Thailand)）
2:30 PM - 2:45 PM

[4-1330-A-06] Quality Characteristics of Thai Coconut Candy as Affected by Rice Starch-Based Film Enriched with Dragon Fruit Peel Extract

*Wantida Homthawornchoo[1,3], Nur Fairuza Syahira Mohamad Hakimi[2,1], Saroat Rawdkuen

[1,3] （1. Food Science and Technology Program, Mae Fah Luang University(Thailand), 2. Food Sciences and Technology Program, Universiti Teknologi MARA(Malaysia), 3. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University(Thailand)）

2:45 PM - 3:00 PM

[4-1330-A-07] Antifungal Packaging for Prolonging Shelf Life of Table Grapes

*SIRIPORN LUESUWAN[1]　（1. MAE FAH LUANG UNIVERSITY(Thailand)）

3:00 PM - 3:15 PM

1:30 PM - 1:45 PM (Wed. Sep 4, 2019 1:30 PM - 3:30 PM Hall A)

[4-1330-A-01] Edible Coatings Control Shrivel and Maintain Quality of Nectarines during Simulated Export Conditions

Shannon Riva[2], *Olaniyi Amos Fawole[1], Umezuruike Linus Opara[1,2]　（1. Postharvest Technology Research Laboratory, South African Research Chair in Postharvest Technology, Department of Food Science, Stellenbosch University(South Africa), 2. Postharvest Technology Research Laboratory, South African Research Chair in Postharvest Technology, Department of Horticultural Science, Stellenbosch University(South Africa)）
Keywords: Weight loss, 'August Red', Shrivel, Postharvest, Respiration rate

Nectarines are considered as one of the most important stone fruits of temperate origin. The fruit's perishable nature, however, limits its commercial success; fruit is often exposed to very long handling chains during export. Edible coatings have shown great potential in maintaining quality and extending shelf life of fresh produce, and thus their application to nectarines is worth investigating as a green postharvest solution. Nectarines ('August Red') were treated with alginate (2%), chitosan (1.5%), gellan gum (0.5%) and gum arabic (2%) coatings and stored at -0.5 ± 1 ˚C, 80 ± 5% RH for 35 days and then at 21 ± 1 ˚C, 95 ± 5% RH for 20 days, simulating the cold storage shipment period and commercial shelf life period, respectively. Fruit were assessed at intervals for weight loss, fruit firmness, and colour changes, as well as respiration rates and ethylene evolution. Physiological disorders such as shrivel occurrence were assessed, as well as decay. The investigated edible coatings showed different effects, with gum arabic having the most effective performance on the plums. Fruit coated with gum arabic had the least weight loss while the highest weight loss was observed in uncoated fruit. In addition, the coating prolonged storage life by delaying fruit ripening and decreasing respiration rate and ethylene production. Furthermore, shrivel incidence was significantly ($p<0.05$) lower in fruit coated with gum arabic (2%) compared to the uncoated fruit (50%). Overall, the results suggested that gum arabic coating of nectarines was most effective to extend the storage life of nectarines ('August Red') and could be investigated further for commercial application.

1:45 PM - 2:00 PM (Wed. Sep 4, 2019 1:30 PM - 3:30 PM Hall A)

[4-1330-A-02] Development and Characterization of Chitosan Film Incorporated with Cashew (*Anacardium Occidentale*) Leaf Extracts

*moooksupang - Liangpanth[1,2,3]　（1. Mae Fah Luang University(Thailand), 2. Thomas Karbowiak(France), 3. Wirongrong Tongdeesoontorn(Thailand)）
Keywords: Cashew leaf extract, Antifungal property, Chitosan film

Chitosan is a material gotten from natural which is a non-toxic, biodegradable, biocompatible substance which contains antimicrobial and antioxidant properties. Normally, chitosan can be dissolved in acid affect the viscous solution that is suitable for making a film. Many studies reported that chitosan film combined with plant extracts showed the synergistic effect to antimicrobial properties. The previous studied found that cashew leaf extracts showed the biological properties such as antioxidant, antimicrobial, analgesic and anti-inflammatory. This research aimed to study the antimicrobial activities of cashew leaf extracted with water and 70% ethanol, and to study properties of chitosan film incorporated with cashew leaf extract. The result showed that ten percent of ethanolic and aqueous extracts could inhibit the growth of *Aspergillus niger*. An

ethanolic extract (CLE) had lower of minimal inhibitory concentration (MIC = 6.25 μg/100μl) than those of aqueous extract (CLAq) (MIC = 12.5 μg/100μl) but showed the same minimal fungal concentration (MFC =25 μg/100μl). For film properties, film solutions were prepared as (i) 2% chitosan, (ii) 2% chitosan + 5% CLE (w/v), (iii) 2% chitosan + 5% CLAq (w/v). All the films were determined color, thickness, barrier properties (WVTR, WVP, OTR) and antifungal activities. The highest values of thickness was obtained by chitosan combined with 5% CLE followed by chitosan mixed with 5% CLAq and chitosan film (control). Moreover, chitosan mixed with 5% CLE also gave lower WVTR and WVP than the other films. Furthermore, only chitosan combined with 5% CLE presented the antifungal property against the growth of *A. niger*. The chitosan film combined with cashew leaf extract could be a new alternative way of natural antifungal package which can be used in food and agricultural produce.

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1330-A] Postharvest/Food Technology and Process Engineering (2)

Wed. Sep 4, 2019 1:30 PM - 3:30 PM Hall A (Main Hall)

[4-1330-A-03] Green Synthesis of Zinc Oxide Nanoparticles from Asiatic Pennywort (*Centella asiatica* L.) and Its Effect on the Rice Starch-Gelatin Composite Film

*Wantida Homthawornchoo[1,2], Suttiporn Pinijsuwan[1,2], Saroat Rawdkuen[1,2]　(1. School of Agro-Industry, Mae Fah Luang University(Thailand), 2. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University,(Thailand))

Keywords: Centella asiatica (L.), , Zinc oxide nanoparticles, Gelatin, Rice starch, Antimicrobial activity

Asiatic pennywort (*Centella asiatica* L.) is a plant that is abundantly available in Thailand. The *Centella asiatica* leaf extract (CAE) is known to possess antioxidant and antimicrobial properties as it contains polyphenols. This allows CAE to act as a reducing agents for a green synthesis of zinc oxide nanoparticles (ZnONPs). The ZnONPs provides a good antimicrobial activity, especially to bio-based packaging films. Therefore, in the objectives of this study were *(i)* to synthesize the ZnONPs using the CAE and *(ii)* to investigate the effects of the ZnONPs incorporation on the physicochemical, thermal, and antimicrobial properties of the rice starch-gelatin composite film. Briefly, ZnONPs were synthesis through the green method using CAE. The shape and the size of the prepared ZnONPs were found to be the rod shape of 100-300 nm in length. The ZnONPs were then added into the rice starch-gelatin composite film at different concentrations (i.e., 0, 0.5, 1, 2, and 3%, w/v). As the ZnONPs increased, the thickness of the developed film with ZnONPs addition were found to increase (Approx. 50-70 mm). The tensile strength (TS) were also increased from 3.49 to 4.63 MPa as well as the increasing of the water vapor permeability. The thermal stability of the developed film was also increased. However, the addition of ZnONPs reduced the elongation at break (EAB) (92.20-37.68%) and the film solubility (67.84 - 30.36%). Furthermore, ZnONPs also altered the color, appearance, transmission, and transparency properties of the prepared films with the crystalline structure presented as confirmed by X-ray diffraction (XRD) analysis. The antimicrobial activity of the rice starch-gelatin film enriched with ZnONPs against Gram-positive bacteria (*S. aureus* and *B. cereus*), Gram-negative bacteria (*E. coli* and *S.* Typhimurium), and fungal (*A. niger* and *C. alatae*) by disc diffusion method were found to be higher as the ZnONPs concentration increased while the rice starch-gelatin film without ZnONPs did not show the inhibition zone. Thus, the developed rice starch-gelatin film with ZnONPs could potentially be used as an antimicrobial packaging film.

Green Synthesis of Zinc Oxide Nanoparticles from Asiatic Pennywort (*Centella asiatica* L.) and Its Effect on the Rice Starch-Gelatin Composite Film

Wantida Homthawornchoo[1, 2,]*, Suttiporn Pinijsuwan[1, 2], Saroat Rawdkuen[1, 2]

[1]Food Science and Technology Program, School of Agro-Industry, Mae Fah Luang University, Thailand
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ABSTRACT

Asiatic pennywort (*Centella asiatica* L.) is a plant that is abundantly available in Thailand. The *Centella asiatica* leaf extract (CAE) is known to possess antioxidant and antimicrobial properties as it contains polyphenols. This allows CAE to act as a reducing agents for a green synthesis of zinc oxide nanoparticles (ZnONPs). The ZnONPs provides a good antimicrobial activity, especially to bio-based packaging films. Therefore, in the objectives of this study were *(i)* to synthesize the ZnONPs using the CAE and *(ii)* to investigate the effects of the ZnONPs incorporation on the physicochemical, thermal, and antimicrobial properties of the rice starch-gelatin composite film. Briefly, ZnONPs were synthesis through the green method using CAE. The shape and the size of the prepared ZnONPs were found to be the rod shape of 100-300 nm in length. The ZnONPs were then added into the rice starch-gelatin composite film at different concentrations (i.e., 0, 0.5, 1, 2, and 3%, w/v). As the ZnONPs increased, the thickness of the developed film with ZnONPs addition were found to increase (Approx. 50-70 μm). The tensile strength (TS) were also increased from 3.49 to 4.63 MPa as well as the increasing of the water vapor permeability. The thermal stability of the developed film was also increased. However, the addition of ZnONPs reduced the elongation at break (EAB) (92.20-37.68%) and the film solubility (67.84 - 30.36%). Furthermore, ZnONPs also altered the color, appearance, transmission, and transparency properties of the prepared films with the crystalline structure presented as confirmed by X-ray diffraction (XRD) analysis. The antimicrobial activity of the rice starch-gelatin film enriched with ZnONPs against Gram-positive bacteria (*S. aureus* and *B. cereus*), Gram-negative bacteria (*E. coli* and *S.* Typhimurium), and fungal (*A. niger* and *C. alatae*) by disc diffusion method were found to be higher as the ZnONPs concentration increased while the rice starch-gelatin film without ZnONPs did not show the inhibition zone. Thus, the developed rice starch-gelatin film with ZnONPs could potentially be used as an antimicrobial packaging film.

Keywords: *Centella asiatica* (L.), Zinc oxide nanoparticles, Gelatin, Rice starch, Antimicrobial activity

2:15 PM - 2:30 PM (Wed. Sep 4, 2019 1:30 PM - 3:30 PM Hall A)

[4-1330-A-04] Combination of high pressure processing and heat treatment on quality and antioxidant activity of fresh-cut persimmon

Paweena Jarungjitaree[1], Matchima Naradisorn1[1,2], Daisuke Hamanaka [3], *Sutthiwal Setha[1,2] （1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, 57100, Thailand(Thailand), 2. Research Group of Postharvest Technology, Mae Fah Luang University, Chiang Rai, 57100, Thailand(Thailand), 3. Faculty of Agriculture, Kagoshima University, Kagoshima, 8900065, Japan(Japan)）
Keywords: Antioxidant activity, Browning, Heat treatment, High pressure processing, Persimmon

The effect of high pressure processing (HPP) in combination with heat treatment on storage quality and antioxidant activity of fresh-cut persimmon were investigated. Flesh persimmon (*Diospyros kaki* L., cv. ‘Hiratanenashi’) were cut and treated by HPP at 80 MPa combined with heat treatment at 40°C for 5, 10 and 15 minutes, non-treated was used as a control treatment. Then, they were stored at 5 °C for 8 days. The quality determinations were measured for respiration rate, weight loss, color, flesh firmness, total soluble solid, polyphenol oxidase activity (PPO), total phenolic content (TPC) and antioxidant activity by 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity and ferric reducing antioxidant power (FRAP) assays. The result showed that HPP-treated had significantly lower respiration rate and weight loss than the non-treated. During the period evaluated, L^*, C^* and flesh firmness were significantly lower in HPP-treated than the non-treated. PPO activity increased during storage however HPP-treated for 5 and 10 min could reduce the PPO activity when compared with other treatments. TPC, DPPH radical scavenging activity and FRAP activity decreased during storage but HPP-treated for 5 min maintained significantly higher values of TPC, DPPH and FRAP than the other treatments. HPP (80 MPa) combined with mild heat treatment (40°C) for 5 min provides potentially beneficial maintained high antioxidant activity and reduced respiration rate, weight loss and PPO activity with only a slight translucency caused by high pressure in fresh-cut persimmon.

2:30 PM - 2:45 PM (Wed. Sep 4, 2019 1:30 PM - 3:30 PM Hall A)

[4-1330-A-05] Postharvest Ethylene Application Influences Biochemical Quality of Pummelo Fruit Under Low Temperature Storage

*Paemika Promkaew[1], Varit Srilaong[2], Satoru Kondo[1] （1. Graduate School of Horticulture, Chiba University(Japan), 2. Division of Postharvest Technology, School of Bioresources and Technology, King Mongkut's University of Technology Thonburi (Bangkhuntien)(Thailand)）
Keywords: Ethylene, Pummelo, Low temperature

The effects of exogenous ethylene on the biochemical quality of 'Tumtim Siam' pummelo (*Citrus maxima* Burm.) fruit under low temperature were investigated. Fruit were treated with 10 µL L^{-1} ethylene for 12 hours at 25 °C and then stored at 10 °C for 40 d. Fruit treated with ethylene showed a remarkable ethylene production after 10 d and respiration rate at 20 d. The chlorophyll concentrations and hue angle value of peel and pulp were decreased by ethylene treatment compared to the untreated control. The carotenoid, phenolic and ascobic acid concentrations were significantly higher at 20 and 30 d in the ethylene treated-fruit than the untreated control. However, sugar concentrations were significantly higher at 20 and 30 d in the untreated control than the fruit treated with ethylene. Overall, ethylene had a significant effect on the

peel and pulp coloration and on the biochemiacl qualities in pulp of the fruit during storage at low temperature.

Oral Session | Postharvest/Food Technology and Process Engineering
[4-1330-A] Postharvest/Food Technology and Process Engineering (2)
Wed. Sep 4, 2019 1:30 PM - 3:30 PM Hall A (Main Hall)

[4-1330-A-06] Quality Characteristics of Thai Coconut Candy as Affected by Rice Starch-Based Film Enriched with Dragon Fruit Peel Extract

*Wantida Homthawornchoo[1,3], Nur Fairuza Syahira Mohamad Hakimi[2,1], Saroat Rawdkuen[1,3] (1. Food Science and Technology Program, Mae Fah Luang University(Thailand), 2. Food Sciences and Technology Program, Universiti Teknologi MARA(Malaysia), 3. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University(Thailand)）
Keywords: Rice starch-based film, Dragon fruit peel extract, Coconut milk candy, Lipid oxidation, Antioxidant

Thai coconut candy or *Kalamae*, a soft and luscious caramel candy, typically made from glutinous rice flour, palm sugar, and coconut milk. The Thai coconut candy short shelf life, due to lipid oxidation and starch retrogradation of the Thai coconut candy, has affected its quality and subsequently limited its market growth. In addition, the use of bio-based plastic instead of petroleum-based plastic packaging is now a good practice as the latter is harmful to the environment and sea life. So, the objectives of this study are *i)* to apply the starch-based (RS) film with dragon fruit peel extract (DPE) as a packaging of the Thai coconut candy and *ii)* to determine the quality attributes of the Thai coconut candy as affected by the RS-DPE film. The dragon fruit peel was extracted and freeze-dried to obtain the DPE powder. The rice starch-based film with DPE of 2 %w/w solid was prepared by the air-dried casting method. The thickness, color, appearance, water vapor permeability (WVP), total phenolic content (TPC), total betacyanin content (TBC), and DPPH scavenging activity (%DPPH) of the RS-DPE film were determined. Film appearance was smooth. The lightness (*L**) of the RS-DPE film, as compared to the RS film without DPE, was lowered but *a** and *b** values were increased toward redness and yellowness, respectively. The thickness of the film was also increased while the WVP was not altered. The RS-DPE film exhibited a significantly higher amount of TPC and TBC with the higher %DPPH than the RS film. The Thai coconut candies were wrapped in the RS-DPE films and kept in the control chamber at 25 ºC, 50% RH. The commercial polypropylene plastic was used as a control. The quality characteristics of the Thai coconut candy were assessed at day 1, 3, 5, 7, and 9, respectively. There was no significant difference ($p < 0.05$) in the a_w and moisture content (MC) of the candies packaged in both film treatments. As the storage time progressed, the aw and MC were reduced. The results were confirmed by the significant increase in hardness (N) and springiness (%) of the candies wrapped in both film types as time increased. However, hardness and springiness of the candies wrapped in the RS-DPE film were higher than that packed in the commercial film. The lipid oxidation expressed as Thiobarbituric acid reactive substance (TBARS) was increased in all treatments over time. However, the candies wrapped in RS-DPE film showed a significant lower in TBARS values. Thus, this could be implied that the RS-DPE film helps delay lipid oxidation in the candy but the water barrier needed to be improved in order to retain the soft and springy texture of the Thai coconut candy.

Quality Characteristics of Thai Coconut Candy as Affected by Rice Starch-Based Film Enriched with Dragon Fruit Peel Extract

Wantida Homthawornchoo[1, 3,]*, Nur Fairuza Syahira Mohamad Hakimi[1, 2], Saroat Rawdkuen[1, 3]

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[2]Food Sciences and Technology Program, School of Applied Science, Universiti Teknologi MARA, Malaysia
[3] Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University, Thailand

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ABSTRACT

Thai coconut candy or *Kalamae*, a soft and luscious caramel candy, typically made from glutinous rice flour, palm sugar, and coconut milk. The Thai coconut candy short shelf life, due to lipid oxidation and starch retrogradation of the Thai coconut candy, has affected its quality and subsequently limited its market growth. In addition, the use of bio-based plastic instead of petroleum-based plastic packaging is now a good practice as the latter is harmful to the environment and sea life. So, the objectives of this study are *i)* to apply the starch-based (RS) film with dragon fruit peel extract (DPE) as a packaging of the Thai coconut candy and *ii)* to determine the quality attributes of the Thai coconut candy as affected by the RS-DPE film. The dragon fruit peel was extracted and freeze-dried to obtain the DPE powder. The rice starch-based film with DPE of 2 %w/w solid was prepared by the air-dried casting method. The thickness, color, appearance, water vapor permeability (WVP), total phenolic content (TPC), total betacyanin content (TBC), and DPPH scavenging activity (%DPPH) of the RS-DPE film were determined. Film appearance was smooth. The lightness (*L**) of the RS-DPE film, as compared to the RS film without DPE, was lowered but *a** and *b** values were increased toward redness and yellowness, respectively. The thickness of the film was also increased while the WVP was not altered. The RS-DPE film exhibited a significantly higher amount of TPC and TBC with the higher %DPPH than the RS film. The Thai coconut candies were wrapped in the RS-DPE films and kept in the control chamber at 25 ºC, 50% RH. The commercial polypropylene plastic was used as a control. The quality characteristics of the Thai coconut candy were assessed at day 1, 3, 5, 7, and 9, respectively. There was no significant difference ($p < 0.05$) in the a_w and moisture content (MC) of the candies packaged in both film treatments. As the storage time progressed, the aw and MC were reduced. The results were confirmed by the significant increase in hardness (N) and springiness (%) of the candies wrapped in both film types as time increased. However, hardness and springiness of the candies wrapped in the RS-DPE film were higher than that packed in the commercial film. The lipid oxidation expressed as Thiobarbituric acid reactive substance (TBARS) was increased in all treatments over time. However, the candies wrapped in RS-DPE film showed a significant lower in TBARS values. Thus, this could be implied that the RS-DPE film helps delay lipid oxidation in the candy but the water barrier needed to be improved in order to retain the soft and springy texture of the Thai coconut candy.

Keywords: Rice starch-based film, Dragon fruit peel extract, Coconut milk candy, Lipid oxidation, Antioxidant,

3:00 PM - 3:15 PM (Wed. Sep 4, 2019 1:30 PM - 3:30 PM Hall A)

[4-1330-A-07] Antifungal Packaging for Prolonging Shelf Life of Table Grapes

*SIRIPORN LUESUWAN[1]　(1. MAE FAH LUANG UNIVERSITY(Thailand)）

Keywords: Table grape, Essential oil, Antifungal packaging, Pathogenic fungi

The table grape (*Vitis vinifera* cv. 'Beauty seedless') becomes popular fruit in Thailand because it's seedless and sweet. The fungal decays in table grapes from *Botrytis cinerea*, *Penicillium* spp., *Aspergillus* spp., and *Rhizopus stolonifera* (soft-rot mold) are the main postharvest problems. To dissolve this problem for grapes exportation and distribution, many countries usually used sulfur dioxide (SO_2) fumigation in sachet and pad forms for inhibiting the fungal growth. However, the sulfur dioxide fumigation is not accepted in many countries because sulfur dioxide residues are potentially hazardous to human health. So, this research was studied about the new antifungal packaging for delaying the mold growth in table grapes by using natural essential oils. The concentration of essential oils (clove, cinnamon, thyme, peppermint, lemon, bergamot, ginger, spearmint, and lemongrass oils) at 0.5, 1, 2 and 5% (w/v) were used for screening the antifungal property against *Aspergillus* spp. by using the disk diffusion method, minimal inhibitory concentration (MIC) assay, minimal fungicidal concentration (MFC) assay, and determination of radial growth. The results showed that the clove, cinnamon and lemongrass oils could inhibit the growth of *Aspergillus* sp. In part of sensory evaluation by untrained panelists, they were accepted the odor of clove oil when apply in grape. So, clove oil was selected to use in application part. In application, grapes were packed into perforated polypropylene (PP) bag and devided into 5 groups; (i) without essential oil (control), (ii) with the essential oil in absorbent pad, (iii) with oriented polypropylene (OPP) film coated 7% polyvinyl alcohol mixed with essential oil, (iv)with OPP film coated 7% polyvinyl alcohol mixed with essential oil and 1% halloysite clay, and (v) commercial sulfer dioxide pad. The 300 grams of table grapes were packed and stored at 14˚C, 75 %RH for 30 days. The weight loss and sensory evaluation of table grapes were determined every 3 days. The results showed that all of treatments could delay the growth of the pathogenic fungi except control. The OPP film coated 7% polyvinyl alcohol mixed with essential oil with and without halloysite clay could prolong shelf life of table grapes for 21 days. While control, perforated PP bag with the essential oil absorbent pad and commercial pad could prolong shelf life of table grape for 12, 15, and 18 days respectively.

Oral Session | Food Function/Nutrition

[4-1330-C] Functional/Wellness Foods &Nutrition (1)

Chair:Rosires Deliza(Embrapa Food Technology, Brazil)
Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room C (3rd room)

[4-1330-C-01] **The Influence of The Front-of-Pack Nutrition Labelling Schemes on Helping Healthier Food Choices by Consumer**

*Rosires Deliza[1], Marcela Alcantara[2], Renata Vaqueiro Pereira[3], Gastón Ares[4] （1. Embrapa Food Technology(Brazil), 2. PDJ_CNPq/Embrapa Food Technology(Brazil), 3. Federal Rural University of Rio de Janeiro(Brazil), 4. Universidad de la Repú blica(Uruguay)）

1:30 PM - 1:45 PM

[4-1330-C-02] **Colour and Chemical Composition of Karasumi-like Chinook salmon (*O. tshawytscha*) Roe Relevant to its Quality**

*Senni Bunga[1,3], John Birch[1], Alan Carne[2], Alaa El-Din A Bekhit[1] （1. Departement of Food Science, University of Otago, New Zealand(New Zealand), 2. Department of Biochemistry, University of Otago, New Zealand(New Zealand), 3. Indonesia Endowment for Education (LPDP), Indonesia(Indonesia)）

1:45 PM - 2:00 PM

[4-1330-C-03] **Effect of Inulin and *Carissa carandas* L. Supplementation on Physicochemical and Microbiological Properties of Frozen Yogurt**

*Kamonwan Manowan[1,2], Ni-orn Chomsri[1,2] （1. Agricultural Technology Research Institute, Rajamangala University of Technology Lanna(Thailand), 2. Faculty of Sciences and Agricultural Technology, Rajamangala University of Technology Lanna(Thailand)）

2:00 PM - 2:15 PM

[4-1330-C-04] **Utilization of Banana Agricultural Waste: Effects of Processing Conditions on Properties of Unripe Banana (*Musa Cavendish*) Pulp and Peel Flours**

*Natthawuddhi Donlao[1,2], Asia Perin[1], Nasuha Bunyameen[1] （1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 2. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University, Thailand(Thailand)）

2:15 PM - 2:30 PM

1:30 PM - 1:45 PM (Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room C)

[4-1330-C-01] The Influence of The Front-of-Pack Nutrition Labelling Schemes on Helping Healthier Food Choices by Consumer

*Rosires Deliza[1], Marcela Alcantara[2], Renata Vaqueiro Pereira[3], Gastón Ares[4] （1. Embrapa Food Technology(Brazil), 2. PDJ_CNPq/Embrapa Food Technology(Brazil), 3. Federal Rural University of Rio de Janeiro(Brazil), 4. Universidad de la República(Uruguay)）
Keywords: Nutritional Warnings, Guideline Daily Amounts, Front-of-pack, Nutrition information, Food policy

Several new graphic designs for FOP nutrition labelling schemes are being developed worldwide; therefore, it is necessary to get an understanding of how they can influence the efficacy to facilitate the identification of nutrients associated with non-communicable diseases (NCDs). In the present work five nutritional warnings (black and red magnifier, black octagon, black triangle and red circle) that are being considered by Brazilian national authorities for implementing in the country, was compared with two of the most studied schemes (the guidelines daily amounts (GDA) and the traffic-light system) in terms of the perception of the product healthfulness by consumer. Fictitious labels of eight product categories frequently consumed in Brazil were considered: sponge cake, orange nectar, frozen lasagna, cereal bar, breakfast cereal, chocolate flavoured milk, yogurt and savoury snack. For this, an online survey with 1932 participants from the five regions of the country was carried out to evaluate their ability to use FOP nutrition labelling schemes to correctly identify the most healthful product in a set, as well as the high nutrient content in a product. In addition, the influence of FOP nutrition labelling schemes on perceived healthfulness was evaluated. Finally, consumers' perception of the schemes was gathered using an open-ended question. As expected, the GDA system showed the worst performance in terms of consumers' ability to identify the most healthful product in a set, and to identify products with high content of nutrients associated with NCDs. All the schemes that included textual or graphical interpretation aids showed a similar performance in improving consumers' ability to identify the most healthful product in a set of alternatives. However, the schemes based on familiar marks (the traffic-light, red circle, black octagon and black triangle) tended to show better performance compared to the red and black magnifier. Consumers showed a positive attitudes towards all the FOP nutrition labelling schemes, in line with previous studies that have highlighted that this public policy is strongly supported by citizens.

Oral Session | Food Function/Nutrition
[4-1330-C] Functional/Wellness Foods &Nutrition (1)
Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room C (3rd room)

[4-1330-C-02] Colour and Chemical Composition of Karasumi-like Chinook salmon (*O. tshawytscha*) Roe Relevant to its Quality

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Keywords: Salmon roe, fermentation, colour, chemical composition

Salt drying of fish products has been used as a food preservation method since ancient times for the production of shelf-stable products. Chinook salmon (*O. tshawytscha*) fish roe was used to produce a salted-dried (Karasumi-like) roe product. The changes in colour properties and chemical compositions (proximate, pH, water activity, salt content, and acidity value) were studied over 20 days of salt-drying at 4°C. Results showed that redness (a*) and yellowness (b*) were decreased gradually in value from 15.76 (at day 0) to 23.88 (at day 20) and from 24.88 (at day 0) to 3.44 (at day 20), respectively. Protein, lipid, ash, carbohydrate, salt content, and acidity value were higher ($P < 0.05$) after salted-drying. The water activity decreased from 0.98 (at day 0) to 0.82 (at day 20). The pH value fluctuated but was overall lower at the end of the processing step. This study highlighted the potential for converting raw or frozen salmon roe into a salted-dried product with added value, that may have beneficial effects on the nutritional and functional characteristics of the processed product.

Colour and chemical composition of karasumi-like Chinook salmon (*O. tshawytscha*) Roe Relevant to its Quality

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ABSTRACT

Salt drying of fish products has been used as a food preservation method since ancient times for the production of shelf-stable products. Chinook salmon (*O. tshawytscha*) fish roe was used to produce a salted-dried (Karasumi-like) roe product. The changes in colour properties and chemical compositions (proximate, pH, water activity, salt content, and acidity value) were studied over 20 days of salt-drying at 4°C. Results showed that redness (a*) and yellowness (b*) were decreased gradually in value from 24.49 (at day 0) to 5.18 (at day 20) and from 24.88 (at day 0) to 3.44 (at day 20), respectively. Protein, lipid, ash, carbohydrate, salt content, and acidity value were higher ($P < 0.05$) after salted-drying. The water activity decreased from 0.98 (at day 0) to 0.82 (at day 20). The pH value fluctuated, but was overall lower at the end of the processing step. This study highlighted the potential for converting raw or frozen salmon roe into a salted-dried product with added value that may have beneficial effects on the nutritional and functional characteristics of the processed product.

Key words: Salmon roe, fermentation, colour, chemical composition.

1. INTRODUCTION

Fish roe refers to the eggs of fish is widely consumed throughout the world as delicacy food. Chinook salmon (also known as king salmon) is the most important fish species farmed commercially in New Zealand. According to the New Zealand Aquaculture Industry (2018) reported harvests of approximately 8,000 metric tonnes of salmon and contributed more than 50% of the world's total king salmon supply. Large scale of the production leads to the utilization of the seafood by-product such as fish viscera, skin, head, and dark meat into new products such as food, nutraceuticals, pharmaceuticals and biotechnological applications (Kim & Mendis, 2006). The mature roe of Chinook salmon accounts for approximately 200 – 400 g of the total body weight and is considered a low-value by-product of the seafood industry.

Fish roe is rich in proteins, vitamins, minerals and ω-3 polyunsaturated fatty acids, but the content of these nutrients vary amongst fish species (Bledsoe et al., 2003). Information on the nutrition values of fresh or frozen fish roe has been reported widely in previous studies (Al-Sayed Mahmoud et al., 2008; Bah et al., 2016; Bekhit et al., 2009b; Heu et al., 2006; Intarasirisawat et al., 2011; Mol & Turan, 2008), however, little information about changes in their composition upon processing. Processing may play an important role to improve the nutritional values of fish roe or it may degrade some components that might be beneficial for human health. Fish roe products are available in different forms. Salted, salted-dried, and salted-fermented roe are some of the roe products that are consumed in certain countries. In Asian countries, such as in Japan, South Korea and Taiwan, fish roe from salmon, pollock, flying fish, and herring are salted and known as Ikura, Tarako, Tobiko, and Kazunuko, respectively. Mullet fish species have also been the main roe used in a salted-dried product, which is known in Japan as Karasumi; Avgotaraho in Greece; Bottargo in Italy and Batarekh in Egypt. Salted-fermented fish roe is known as Karashi-mentaiko in Japan and Jeotgal in Korea.

In this study, Chinook salmon roes were used to produce a salted-dried roe product similar to Karasumi. Changes in colour and physicochemical properties (roe skein size, proximate composition, pH, acidity, salt content, water activity, and weight loss) during the salted-drying process are important, as these factors will influence the quality and consumer's perceptions of the fish roe.

2. Material and Methods

2.1. Fish roe sample and processing

Fresh grade 1 Chinook salmon (*O. tshawytscha*) roe was used in this current study. The weight, length, and width of the roes were measured before processing. The surface of the roe skein then was covered with sea salt (7% of the wet weight). The drying process was conducted at 4°C in a chiller room for 20 days at fixed relative humidity of 80%. For optimal air circulation, stainless steel wire racks were

used for the drying of the roe samples. Sampling of the roe was carried out every 4 days. Each sampling day, three whole fish roes were randomly chosen for further analysis. Schematic overview of the roe processing is shown in Figure 1.
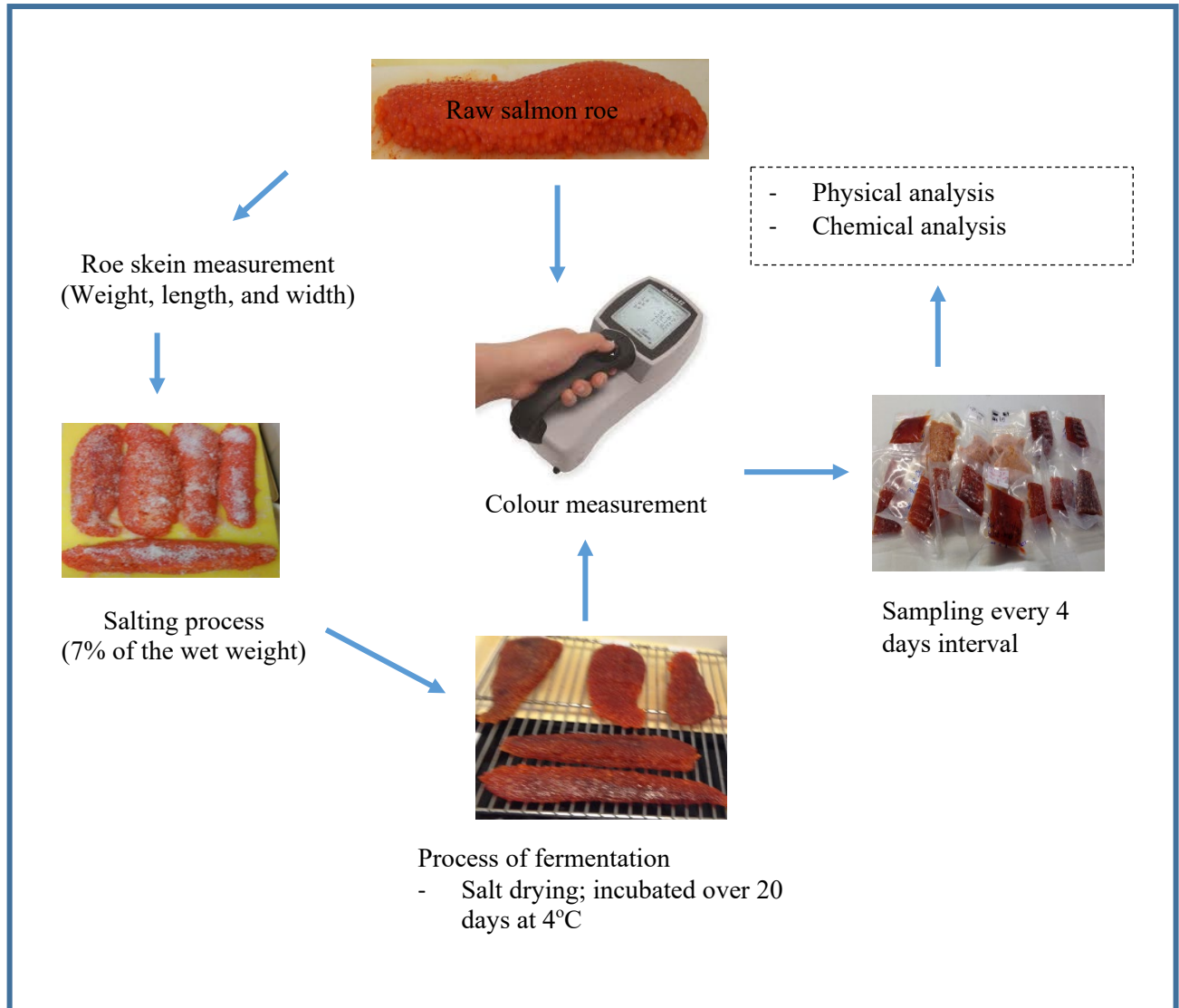

Figure 1. Diagram process of salted-dried product from raw roe salmon

2.2. Measurement of roe colour

The surface colour of the roes was measured using a Hunterlab MiniScan XE Plus Model 45/0-L (Hunter Associates Laboratory, Inc., Reston, VA, USA). The colour parameters L*, a*, b*, representing lightness, redness/greenness, and yellowness/ blueness, respectively, were measured at each sampling time. The instrument was calibrated using a standard black glass and a white tile according to the manufacture

guidelines. The sample was placed on the tray and covered with a polyvinyl film (AEP FilmPac, Ltd, New Zealand). Three replicate readings from random positions were taken from each skein and the average value was used for statistical analysis. Chroma and hue angle were calculated based on the following formula: C= [a*2 + b*2]1/2 and HA=tan-1 b*/a*, respectively.

2.3. Chemical analysis

2.3.1. Weight loss

Weight loss was measured every 4 days over the 20 days of salt-drying by removing the individual roe from the chiller room and were weighed using a Sartorius Type Universal U 4600 P scale.

2.3.2. Proximate compositions

Moisture content was determined using a gravimetric measurement of water content by freeze-drying of 5 g of roe sample until a constant weight was achieved. Total lipid was extracted from 3 g of freeze-dried roe using Soxhlet extraction according to the official method (AOAC, 1995). Crude protein content was determined by Kjeldahl-Nitrogen method (% N*6.25) according to the AOAC Official method 981.10 using a Tecator Kjeltec Auto Sampler System 1035 apparatus. Ash content was determined on 2 g of freeze-dried samples, carried out in duplicates from three separated skeins. Carbohydrate content was calculated by subtracting the sum of lipid, protein, ash and moisture content from the total weight of samples. Energy value was determined using the following formula as described by Falch et al. (2010):

[Energy (kcal/100g) = (lipid * 9) + (protein * 4) + ((carbohydrate * 4)]

2.3.3. pH and total acidity

The pH value was measured using a pH meter (HANNA Instrument model PH209, USA) in a 10 g of sample homogenized with 100 mL of deionized water at 6000 rpm using a homogenizer (IKA® T25 Digital Ultra Turrax, TLS, Total Lab-System, Ltd) and the mixture was filtered. For titratable acidity, using the same homogenates, 1 ml of the roe filtrate mixed with 99 ml of deionized water then was determined by titrating against 0.1N standard sodium hydroxide (NaOH) solution. Complete titration was achieved when the colour turned to pink against three drops of phenolphthalein indicator. The % (v/v) acid in the sample was calculated as lactic acid using the following formula:

$$Acidity = \frac{0.009 \ x \ ml \ of \ 0.1 \ N \ NaOH \ x \ F \ x \ 100}{Sample \ (ml)}$$

2.3.4. Salt content

Salt content of the roe product was determined using the titrimetric method according to the AOAC (1995). Preparation of the sample for the chloride ion concentration was done by placing 1 ml of roe filtrate (prepared for acid value determination) into a 250 ml of Erlenmeyer flask, and made up to a final volume of 100 ml with deionized water. Small quantities of $NaHCO_3$ were added in order to adjust the pH of the solution to 7 and a 2 ml of potassium chromate was added to the mixture. The mixed solution then was

titrated with 0.1M of standard silver nitrate solution to the first permanent appearance of red-brown Ag_2CrO_4 achieved.

2.3.5. Water activity

Water activity was measured at 25^oC using a water activity meter (Aqualab Dew Point Model 4TE) on fish roe product before freeze-drying. The analysis was done according to the manufacturer instructions.

3. RESULTS AND DISCUSSION

3.1. Roe skeins of the weight and size

Individual roe skeins had an average weight of 301.31 ± 63.70 g (ranged 200.9 to 422.5 g), length 25.75 ± 3.11cm (ranged from 20.0 to 30.0 cm), and the width 6.21 ± 1.31 cm (ranged between 4.5 and 9.0 cm). In this study, only whole skeins (intact and un-damaged) roes were used, the size of fish roe can be very important consideration that could influence the processing conditions (e.g. salting, drying/fermentation times, salt penetration rate) and suitability of the fish roes for certain products (e.g. product like caviar, salted-, dried- or fermented roe products).

Table 1. Average (mean ± SD) values of weight and size (length and width) of the skeins of fresh salmon roe used in the present research.

Parameter	Mean ± SD	Range
Weight (g)	301.31 ± 63.70	200.9 – 422.5
Length (cm)	25.75 ± 3.11	20.0 – 30.0
Width (cm)	6.21 ± 1.31	4.5 – 9.0

3.2. Colour of the raw roes and salted-dried roe product

Colour and appearance attract the consumers to a product and can support marketing of the product. Colour parameters characterized as lightness (L*), redness (a*), yellowness (b*), chroma (C*), and hue (h*) for raw and processed (salted-dried) roes are shown in Table 2. During 20 days of drying, the roe reduced the colour from red-pink (day 0) to dark red-brown (after day 20 of processing). The a* value decreased from a 24.49 ± 3.27 (at day 0) to a 5.18 ± 0.95 (at day 20), also to the b* value decreased from a 24.88 ± 2.01 (at day 0) to a 3.44 ± 0.96 (at day 20). However, the L* value decreased with the progression of salt drying periods up to day 8 from an average of 45.51 ± 4.31 (at day 0) to 33.00 ± 6.20 (at day 8). Then, followed by the rising up to 39.58 ± 3.19 at the end of salt drying time. The migration of moisture, salt uptake during incubation could affected pigments in the roe, leading to reduction in the redness and chroma. According to (Hayabuchi et al., 1997), processing condition, such as salting, generally controls the colour of the final product due to chemical changes and reduction of moisture. Studies by Bekhit et al. (2009a) on six different New Zealand fish species and Bekhit et al. (2018) on hoki roe found that processing (e.g. fermentation) at different fish species, temperatures and times could affect the microbiological status

and the physicochemical properties of the final product, including colour, as these factors play important part in streamlining quality procedures.

Table 2. Colour changes in salted-dried salmon roe processed at 4ºC.

Fermentation days	L* (Lightness)	a* (Redness)	b* (Yellowness)	C* (Chroma)	HA* (Hue angle)
0	45.51±4.31	24.49±3.27	24.88±2.01	34.99±2.82	45.59±4.36
4	27.45±1.18[d]	15.45±2.09[b]	11.25±2.02[b]	19.14±2.75[b]	35.95±2.79[bc]
8	25.95±3.31[d]	9.75±2.02[c]	7.69±2.30[c]	12.46±2.90[c]	37.84±3.88[b]
12	33.00±6.20[c]	12.67±1.92[bc]	9.49±1.69[bc]	15.84±2.46[bc]	36.752±2.51[bc]
16	38.66±3.05[b]	5.75±1.54[d]	3.71±1.35[d]	6.86±2.00[d]	32.13±3.73[c]
20	39.58±3.19[b]	5.18±0.95[d]	3.44±0.96[d]	6.23±1.27[d]	33.25±4.12[bc]

Each value is the mean ± standard deviation of measurements in three individual roe skeins (n=3).
[a-d] Values in the same column with different superscript are significantly different ($P<0.05$).

3.3. Chemical composition

3.3.1. Proximate

Table 3 gives the effect of salt drying during fermentation time over 20 days on the chemical compositions of the raw salmon roes compared to the salted-dried roes. The moisture content was reduced significantly ($P<0.05$) by about 30% due to the salting out and drying effects from 56.41 ± 0.57% (at day 0) to 25.44 ± 2.72% (at day 20). The loss in moisture was accompanied by increases in ash content, which reflect the increase in salt concentration as well as the apparent increase due to drying process. The ash content increased significantly ($P<0.05$) from 2.34 ± 0.29% at zero time to 6.70 ± 1.11% at the end of drying time.

The protein content of the raw roes was 27. 51 ± 0.26 %. At the end of the processing, the protein content of the roe had increased to 40.39 ± 0.21%. The reason for the protein rate being higher is moisture loss, which may be due to increase of dry matter. Rodrigo et al. (1998) when salting and drying the roe from Hake and Ling fish found that the protein content in the final product ranged between 39.1% and 43.6%, respectively. The obtained results are in agreement with the results performed by Rodrigo et al. (1998).

The percentage of lipid content of the salted-dried salmon roes also exhibited an increasing trend during the salted-drying process. The lipid content of the raw roe was 10.30 ± 1.41% and as salt drying time progressed, the final product had a lipid content higher between 12.83 ± 1.83% and 18.05 ± 2.54%. The changes in the lipids content over the processing time is due to the salt-out and salt-in process throughout the roe skein. The apparent increase in lipid content could be due to the decrease in moisture due to the salting effect and the increased osmosis outside the roe skein. The crude lipid contents found in this study was in the range reported by Bledsoe et al. (2003) for salmon roe products which ranged from 8% to 25%. Mol and Turan (2008) also stated that the lipid content found in red salmon roe was 26.8% and

Bekhit et al. (2009b) reported that lipid content of mature and immature chinook salmon roe was 10.6% and 8.4%, respectively.

The carbohydrate content of the salted and dried roes ranged between $3.43 \pm 1.03\%$ and 9.42 ± 1.51 %. In general, the carbohydrate content increased during the process (Bekhit et al., 2018). The increase in carbohydrate may due to the decrease in moisture content due to the salt drying method.

The energy value of the raw roe was 211.78 ± 0.57 kcal/100g. Salt drying increased the energy value to 370.92 ± 16.67 Kcal/100g at the final day of process, which can be attributed mostly to a loss in moisture content and an apparent increase in other components (Bekhit et al., 2018).

3.3.2. pH and acidity value

The pH and acidity value are factors that are used as indicators of degree of freshness or spoilage and have an impact on product safety and flavour during the development of the fermentation (salted-dried and salted-fermented) process (Bekhit et al., 2018). The changes in pH and acidity value of the roe were observed over the 20 days of salted-drying (Table 3). The initial pH of raw roe was 5.74. This value is in agreement with that of Bledsoe et al. (2003) who reported the pH value of the salmon roe products varied between 5.5 and 5.8. Bekhit et al. (2009b) found that the pH of mature salmon roe was 5.62. In this study, pH value remained constant during processing, but a slight decrease to 5.65 was found at the end of processing time. This finding and previous findings might be attributed to from which the roes were obtained and the different treatments or processing applied. According to statistical assessment performed in this current study, the different was significant ($P<0.05$) during salt drying throughout the entire processing period.

The acid value of the samples which are expressed as percent lactic acid, varied ($P<0.05$) with the increasing of salt drying time. At the first 12 days of salt drying, the acid value was $0.18 \pm 0.03\%$ risen from $0.05 \pm 0.01\%$ when the roe was unprocessed. There was a slight reduction in value after day 16 ($0.16 \pm 0.03\%$) and at the final day of processing where the acid value declined to $0.014 \pm 0.02\%$. The fluctuated acid value according to Adams and Hall (1988) implied that the growth of LAB that produces mixtures of organic acids occurred during the process of salt drying.

3.3.3. Salt content

The salt content of raw roe was $0.58 \pm 0.02\%$ and to be $5.29 \pm 1.37\%$ by the 20th day in salt drying process. The increase in the salt content is due to partial dehydration and salt migration to the roes. The salt content has a significant linear increasing effect on the moisture content ($P<0.05$), which gradually reduced during process of salt drying. Mean values of the salt content obtained in the salted-dried salmon roe were between $3.03 \pm 0.15\%$ (at day 4) and $5.29 \pm 1.37\%$ (at the final day of processing). According to Horner

(1997), the salt concentration above 5%, inhibited most of the microorganisms associated with spoilage are halo-phobic.

3.3.4. Water activity

The availability of water (a_w) measured for the raw roe was 0.98 and decreased to 0.82 up to 20 days of salted-drying. According to Hsu and Deng (1980) salting (15% solid salt) of mullet roe resulted in water loss and salt uptake, which depressed the water activity from 0.975 to 0.858. While, water activity increased after desalting due to water absorption and decreased salt content. The decrease in a_w limits favourable condition for the growth of spoilage microorganisms as studied by Labuza and Rahman (2007) which recorded that the water activity below 0.85–0.86 inhibits pathogenic bacteria, while yeast and moulds are more tolerant to a reduced water activity and cannot grow when water activity is 0.62.

Table 3. Physicochemical compositions following cold incubation at 4ºC of salted-dried salmon roe for over 20 days.

d	Fermentation time (days)					
	0	4	8	12	16	20
Moisture (%)	56.41 ± 0.57^a	43.87 ± 4.14^b	38.63 ± 4.01^{bc}	32.18 ± 1.48^{cd}	27.54 ± 1.83^d	25.44 ± 2.72^d
Lipid (%)	10.30 ± 1.41^c	12.83 ± 1.51^{bc}	13.71 ± 1.68^{abc}	17.21 ± 1.33^{ab}	17.23 ± 1.62^{ab}	18.05 ± 2.54^a
Protein (%N x 6.25)	30.16 ± 0.29^d	34.37 ± 1.61^c	36.28 ± 0.84^c	40.38 ± 2.77^b	45.95 ± 0.86^a	44.29 ± 0.23^{ab}
Ash (%)	2.34 ± 0.29^b	6.52 ± 0.65^a	6.27 ± 1.27^a	6.39 ± 1.32^a	5.36 ± 1.51^{ab}	6.70 ± 1.11^a
Carbohydrate (%)	3.43 ± 1.03^b	5.44 ± 2.34^{ab}	8.30 ± 0.91^{ab}	7.40 ± 1.13^{ab}	7.97 ± 3.85^{ab}	9.42 ± 1.51^a
Energy value (Kcal/100g)	211.78 ± 0.57^d	262.3 ± 24.4^c	289.0 ± 19.9^{bc}	331.79 ± 14.94^{ab}	353.11 ± 7.02^a	370.92 ± 16.67^a
pH	5.74 ± 0.01^a	5.73 ± 0.01^{ab}	5.70 ± 0.02^b	5.75 ± 0.01^a	5.71 ± 0.03^b	5.65 ± 0.03^c
Acidity value (%)	0.05 ± 0.01^e	0.09 ± 0.01^d	0.12 ± 0.02^c	0.18 ± 0.03^a	0.16 ± 0.03^b	0.14 ± 0.02^{bc}
Salt content (%)	0.58 ± 0.02^d	3.03 ± 0.15^c	3.67 ± 0.30^{bc}	3.99 ± 0.75^{bc}	4.52 ± 0.46^{ab}	5.29 ± 1.37^a
Water activity (A_w)	0.98 ± 0.01^a	0.90 ± 0.00^b	0.86 ± 0.00^c	0.84 ± 0.02^d	0.83 ± 0.01^{de}	0.82 ± 0.00^e
Weight loss (%)	100.0 ± 0.00	85.31 ± 1.42	76.49 ± 1.78	71.64 ± 1.97	68.61 ± 2.19	67.44 ± 2.32

Each value is the mean ± standard deviation of three replicates (n=3).
[a-e]values in the same row with different superscript are significantly different (P<0.05).

4. CONCLUSION

Present results indicate that application of salted-drying over 20 days resulted in significant colour change, from red-pink (day 0) to dark red-brown (after day 20 of processing). Salted-drying produced higher protein, lipid, ash, carbohydrate, and energy value compared to the unprocessed roe. Salt drying time and moisture content was found to have a good correlation with pH values, acidity, salt content and aw which can contribute to a more in-depth and understanding of the organisms that involved and their roles in process of salt drying in reducing or increasing bioactive compounds.

ACKNOWLEDGMENT

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Oral Session | Food Function/Nutrition
[4-1330-C] Functional/Wellness Foods &Nutrition (1)
Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room C (3rd room)

[4-1330-C-03] Effect of Inulin and *Carissa carandas* L. Supplementation on Physicochemical and Microbiological Properties of Frozen Yogurt

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Keywords: Frozen yogurt , Inulin , Carissa carandas

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Effect of Inulin and *Carissa carandas* L. Supplementation on Physicochemical and Microbiological Properties of Frozen Yogurt

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ABSTRACT

Frozen yogurt is a frozen dessert claimed to confer health benefits due to the remaining viable bacteria. This study investigates the influence of using inulin and *Carissa carandas* L. (CC) as supplements in the frozen yogurt mixture on the properties of frozen yogurts. The samples were examined for their color, melting behavior, chemical composition, lactic acid bacteria count and sensory evaluation. The results showed that increasing CC contents in the mixtures resulted in higher color values of a* and chroma in the frozen yogurts. Inulin and CC supplementation at the higher levels in frozen yogurts showed higher total solid contents ($p \leq 0.05$). CC supplementation in frozen yogurts at higher levels significantly improved phytochemical properties of final products. Analysis of melting behavior revealed that the melting rate was retarded when higher levels of inulin and CC were employed in the frozen yogurt production. The frozen yogurt containing inulin at 5% and CC at 15% showed a slight reduction of a viable lactic acid bacteria (LAB) count of 0.68 log CFU/g after 90 day-storage at -18 ℃. Sensory results indicated a positive effect of inulin and CC supplementation on the sensory attributes of frozen yogurts.

Keywords: Frozen yogurt Inulin *Carissa carandas*

1. INTRODUCTION

Frozen yogurt, one of frozen dessert is a yogurt ice cream and generally recognized as functional food because LAB starter i.e. *Streptococcus thermophilus* and *Lactobacillus bulgaricus* in yogurt is claimed to confer health benefits (Brunning, 2017; Mena and Aryana, 2018; Das et al., 2019; Terpou et al., 2019; Wasilewska et al., 2019). Furthermore, yogurt has been known as versatile bioactivity including immunity stimulation, gastric disorder treatment, pathogen and intestinal infection protection, diarrhea alleviation, lactose utilization improvement (Feng et al., 2008; Patro-Golab et al., 2015; Aryana and Olson, 2017). However, supplementation of functional ingredients into frozen yogurt may increase beneficial properties on the product regarding consumers' awareness for healthier. Thus, a variety of food ingredients and processing aspects have been examined by researchers to improve the quality of frozen yogurts such as addition of prebiotics and fruits possessing unique and specific functions (Isik et al., 2011; Chanasith et al., 2017; Terpou et al., 2019). Among these practices, supplementation of inulin, a natural polysaccharide belongs to the fructan family has been used widely in frozen yogurt, typically in order to act as prebiotics (Kaur and Gupta, 2002; Yang et al., 2018), enhance probiotic survival (Criscio et al., 2010; Pinto et al., 2012) and improve textural properties of the products (Isik et al., 2011; Muzammil et al., 2017).

CC, botanical name, *Carissa congesta* belonging to Apocynaceae family is an evergreen, spiny shrub and found in Malaya, Thailand, India, Napal, Sri Lanka, Myanmar, Malaysia, Philippines, Cambodia, Vietnam and East Africa (Weerawatanakorn and Pan, 2016; Mohammad et al., 2019). CC has been called Namdaeng (red thorn) or Manao Mai Ru Ho in Thailand (Yuenyongphutthakal et al., 2012; Pewlong et al., 2014; Chomsri et al., 2018). The fruit turns from pinkish white to dark red when ripe and can be eaten raw or processed (Chomsri et al., 2018). The interest in CC has increased during the last decade, because of its pharmacological characteristics, e.g. treatment of constipation, diarrhea, stomachic, anorexia, intermittent fever, mouth ulcer, sore throat, syphilitic pain, burning sensation, scabies and epilepsy (Mehmood et al., 2014, Khatun et al., 2017; Bahdane and Pati, 2017; Singh et al., 2018; Virmani et al., 2017; Madhuri and Neelagund, 2019). The prominent functional properties of the

CC berry fruit, e.g. antioxidant properties, anti-inflammatory effect and biocolorant (Weerawatanakorn and Pan, 2016; Sarkar et al., 2018) shed light on its application in food industry.

The aim of this study was the production of functional frozen yogurt by adding inulin and CC. The influence of added inulin and CC on chemical, physical, and sensorial characteristics of produced frozen yogurt was studied. Moreover, the survival of LAB during frozen yogurt storage was evaluated.

2. MATERIALS AND METHODS
2.1 Frozen yogurt preparation
The experiment was carried out at Agricultural Technology Research Institute, Rajamangala University of Technology Lanna, Lampang, Thailand. Two factors of inulin contents at two levels (0 and 5%) and CC berry contents at 2 levels (10 and 15%) were designated to add in the ice cream mixture for the production of frozen yogurt. Yogurt was produced by adding commercial yogurt culture into pasteurized cow's milk. The mixture of milk and starter yogurt culture was incubated at 45 °C for 5 h. Then, inulin, CC berry, whipping cream, skim milk powder, fructose syrup, and stabilizer were blended with the yogurt at the proportion of 1:1 for ice cream mix preparation. The ice cream mixes were aged at 4 °C for 12 hours. Subsequently, four different frozen yogurt treatments: Frozen yogurt 1; inulin content 0% and CC berry content 10% (I0CC10), Frozen yogurt 2; inulin content 0% and CC berry content 15% (I0CC15), Frozen yogurt 3; inulin content 5% and CC berry content 10% (I5CC10) and Frozen yogurt 4; inulin content 5% and CC berry content 15% (I5CC15) were submitted to the ice cream maker (Model Gel M4, Staff Ice System, Italy). Frozen yogurt samples were packed in cups of 30 ml hardened and stored at −18 °C.

2.2 Physicochemical Analysis of frozen yogurt
The total solids in the frozen yogurt mixture were determined by modified method of Nielsen (2017). pH was measured by digital pH meter (Model C831, Belgium). Total acidity was determined by diluting each 5 ml aliquot of sample in 50 ml distilled water and then titrating to pH 8.2 using 0.1 N NaOH (Iland et al., 2000). Titratable acidity was expressed as lactic acid percentage. Total soluble solid content was determined on an Atago hand-held refractometer. Free alpha amino nitrogen (FAN) was quantified by spectrophotometric method (Intaramoree and Chomsri, 2014). Total anthocyanin content was evaluated by the method of Giusti and Wrolstad (2005). The modified method of Spínola et al. (2015) was used to evaluate total phenolic content. The antioxidant activity was determined by modified method of Wongputtisin et al. (2007).

2.3 Melting behavior of frozen yogurts
The melting behavior was measured according to the method proposed by Arbuckle (1986). The melting rate was calculated by taking 25 g frozen yogurt sample and placing it on a wire mesh screen over a graduated cylinder at 25°C. The dripped mass was recorded every 10 minutes for 60 minutes. The melting rate was calculated by dividing the melted sample over time (g/min). The mass of melted ice cream (%w/w) was plotted against time (min). Melting behavior was determined after one week of frozen storage. This analysis was performed in triplicate.

2.4 Microbiological enumeration
For the enumeration of viable cell counts, 10 g samples were collected from each frozen yogurt directly after packing and the sample I5CC15 at various time intervals (0, 60 and 90 days after manufacturing) during storage at −18 °C. The samples were serially diluted in 90 mL sterile Ringer's solution, diluted and plated on MRS medium.

2.5 Sensory analysis
All the panelists were experienced in frozen yogurt or ice cream. A group of 30 panelists took part in this study. Frozen yogurt products fermented for 2 weeks were evaluated for organoleptic quality. The samples of frozen yogurt were served in random order at −18°C in plastic cups identified with a random 3-digit code. The panelists were suggested to rinse their mouths with water between samples. The panelists were asked to rate the products on the 9-point hedonic scale (Meilgaard et al., 2006)

2.6 Statistical analysis
All the experiments were carried out with replications. Analysis of variance (ANOVA) was used to compare mean differences of the samples. Significant differences between treatments were analyzed by Duncan's new multiple range test (DNMRT) at a 0.95 significance level. Values were expressed as the mean of all replicate determinations with standard deviation.

3. RESULTS AND DISCUSSION
3.1 Physicochemical characteristics
Color analysis of the CC frozen ice creams (Figure 1) was performed and recorded as shown in Table 1. There were significant effects of CC and inulin supplementation on a* (redness) chroma and hue values as interaction. Frozen yogurts from CC contents of 15% showed an increase in a* chroma values compared to frozen yogurts from CC contents of 10%. These results suggest that frozen yogurts from CC contents of 15% showed intensively reddish colors compared with frozen yogurts from CC contents of 10%. This result is associated with an amount of red pigments containing in CC (Mohammad and Ding, 2019).

Figure 1. CC frozen ice creams from different supplementation of inulin and CC contents; a) inulin content 0% and CC berry content 10%, b) inulin content 0% and CC berry content 15%, c) inulin content 5% and CC berry content 10% and d) inulin content 5% and CC berry content 15%

Table 1. Color values of CC frozen ice cream from different supplementation of inulin and CC contents

Treatments	L*	a*	b*	chroma	hue
	ns	*	ns	*	*
I0CC10	55.24+0.34	12.79+0.00[b]	1.27+0.01	12.86+0.00[ab]	5.70+0.04[a]
I0CC15	54.70+1.52	14.91+0.00[ab]	0.73+0.17	14.93+0.01[a]	2.80+0.66[b]
I5CC10	54.71+5.44	10.47+1.47[c]	1.98+2.08	10.73+1.82[b]	3.05+0.02[b]
I5CC15	53.70+2.45	15.27+0.61[a]	0.28+0.16	15.27+0.61[a]	1.08+0.65[c]

ns denotes means are not significantly different ($p > 0.05$)
* Means in a column with the different letters represent significant differences ($p \leq 0.05$)
Inulin content 0% and CC berry content 10%; I0CC10, inulin content 0% and CC berry content 15%; I0CC15, inulin content 5% and CC berry content 10%; I5CC10, and inulin content 5% and CC berry content 15%; I5CC15

The effect of inulin and CC supplementation on total solid contents is displayed in Table 2. Inulin and CC supplementation at the higher levels in frozen yogurts led to higher total solid contents ($p \leq 0.05$). A significant increase of the total solid contents in the frozen yogurt samples by the supplementation of inulin correlated to that of Muzammil et al. (2017), where the frozen ice cream containing 6% inulin had the total solid content higher than the frozen ice cream containing 2% inulin. Total solid contents of CC frozen ice creams ranged from 25 and 33%, while Muzammil et al. (2017) and Isik et al. (2011) reported the total solid contents of frozen yogurts in the ranges of 32% and 35%. The CC supplementation affected the total solid content may be explained by the solid parts found in CC berries such as organic acids, sugars, pigments and fibers (Sarkar et al., 2018; Mohammad and Ding, 2019).
The pH and acidity of various treatments did not differ obviously. However, the results demonstrated that frozen yogurts containing 15 % CC had significantly lower pH and higher total soluble solid contents than frozen yogurts containing 10% CC ($p < 0.05$) as shown in Table 2. The natural titratable acidity of the samples depended on the components containing in the CC frozen ice creams, e.g. lactic

acid in the yogurt and organic acids in the CC. The lowest pH and highest total soluble solid content values were observed in the sample containing inulin 5% and CC 15%. This was probably due to the high acid content in the CC fruits as noted by Chomsri et al. (2017).

Table 2. Chemical property of CC frozen yogurts

Factor	TS (%)	pH	TA (%)	TSS (ºBrix)
Inulin content (A)	*	ns	ns	*
0% (I0)	26.03±1.29[b]	3.92±0.06	1.02±0.05	22.25±2.36[b]
5% (I5)	31.61±2.47[a]	3.89±0.09	0.09±0.07	25.66±3.01[a]
CC content (B)	*	*	ns	*
10% (CC10)	27.26±2.30[b]	3.98±0.01[a]	0.95±0.03	21.58±1.74[b]
15% (CC15)	30.38±3.91[a]	3.83±0.03[b]	1.05±0.03	26.33±2.25[a]
A*B	*	*	ns	ns
I0 x CC10	25.16±0.20[d]	3.97±0.02[a]	0.97±0.00	20.16±0.76
I0 x CC15	29.36±0.17[b]	3.86±0.06[b]	1.07±0.01	24.33±0.57
I5 x CC10	26.90±1.37[c]	3.98±0.01[a]	0.93±0.04	23.00±1.00
I5 x CC15	33.86±0.18[a]	3.81±0.01[c]	1.04±0.04	28.33±0.57

ns denotes means are not significantly different (p>0.05)
* Means in a column with the different letters represent significant differences (p≤0.05)
Total solid content; TS, total acidity (%); TA, total soluble solids (ºBrix); TSS, inulin content 0%; I0, inulin content 5%; I5, CC berry content 10%; CC10 and CC berry content 15%; CC15

The concentrations of total anthocyanin, phenolic, and antioxidant activity in frozen yogurts from different supplementation of inulin and CC contents varied from 10.26 to 13.75, 370 to 460 mg/kg and 55.61 to 71.89 % scavenging effect, respectively. The significant effects of CC supplementation on phytochemical values were observed. Frozen yogurts from CC contents of 15% had higher antioxidant activity (p≤0.05) compared to frozen yogurts from CC contents of 10%. The highest antioxidant activity with 32.17 mg ascorbic acid equivalent/100g sample and 49.88 mg Trolox equivalent /100g sample was achieved in the frozen yogurt containing 5% inulin and 15% CC. This could be explained by CC contains high levels of anthocyanins and phenolic compounds (Pewlong et al., 2014; Weerawatanakorn and Pan, 2016; Chomsri et al., 2017; Sarkar et al., 2018). These substances are most typically responsible for the antioxidant activity exhibited in CC frozen yogurts.

Table 3. Phytochemical contents and antioxidant activity of CC frozen yogurts

Factor	TAC (mg/kg)	TPC (mg/kg)	AO (%)	AOA (mg/100g)	AOT (mg/100g)
Inulin content (A)	ns	ns	ns	ns	ns
0% (I0)	12.00±1.98	434±69	62.49±5.35	27.94±2.39	43.36±3.71
5% (I5)	13.09±3.49	418±78	63.75±9.00	32.17±0.62	44.24±6.24
CC content (B)	*	*	*	*	*
10% (CC10)	10.46±0.98[b]	382±55[b]	56.66±1.81[b]	25.34±0.80[b]	39.32±1.25[b]
15% (CC15)	14.64±2.34[a]	471±56[a]	69.58±2.69[a]	31.13±1.21[a]	48.28±1.87[a]
A*B	ns	ns	*	*	*
I0 x CC10	10.26±0.17	394±83	57.71±1.75[c]	25.81±0.78[c]	40.05±1.21[c]
I0 x CC15	13.75±0.82	475±7	67.27±0.30[b]	30.08±0.13[b]	46.68±0.21[b]
I5 x CC10	10.67±1.50	370±19	55.61±1.36[c]	24.87±0.61[c]	38.59±094[c]
I5 x CC15	13.09±3.49	460±88	71.89±1.45[a]	32.17±0.62[a]	49.88±1.00[a]

ns denotes means are not significantly different (p>0.05)
* Means in a column with the different letters represent significant differences (p≤0.05)
Total anthocyanin content (mg/kg); TAC, total phenolic content (mg GAE/kg); TPC, antioxidant activity (% scavenging effect); AOA, inulin content 0%; I0, inulin content 5%; I5, CC berry content 10%; CC10 and CC berry content 15%; CC15

3.2 Melting behavior of frozen yogurts

Melting is an important assessment criteria of ice cream quality. Many factors influences melting of ice cream, e.g. total solid content, size of the ice particles, amount and size of the fat particles, composition in the mixture, stabilization, presence of air in ice cream (Clarke, 2004; Alfaro et al., 2014; Moriano and Alamprese, 2017, Goral et al., 2018). The melting behavior of the frozen yogurts is presented in Figure 2. The dripping of the first drop of melted frozen yogurts containing inulin required 8-10 minutes, whereas melting time of the first drop of frozen yogurts without inulin supplementation was appeared at less than 5 minutes. In addition, higher levels of inulin in CC frozen yogurts reduced melting rates as obviously seen in meltdown curves. The highest melting rate was recorded for the sample with inulin 5% and CC 15%, whereas the lowest melting rate was obtained from the sample with inulin 0% and CC 10%. Similar results were also reported by Akbari et al. (2016) Junyusen et al. (2017), Rezaei et al., (2014) and Isik et al. (2011). Goral et al. (2018) reported melting time of the first drop ranged between 16-25 minutes in coconut milk-based ice cream with different stabilizer addition, whereas Isik et al. (2011) observed the first drop that frozen yogurts added inulin and isomalt between 8.5-13.5 minutes. This means that components in frozen yogurts substantially affects the melting behavior of the products.
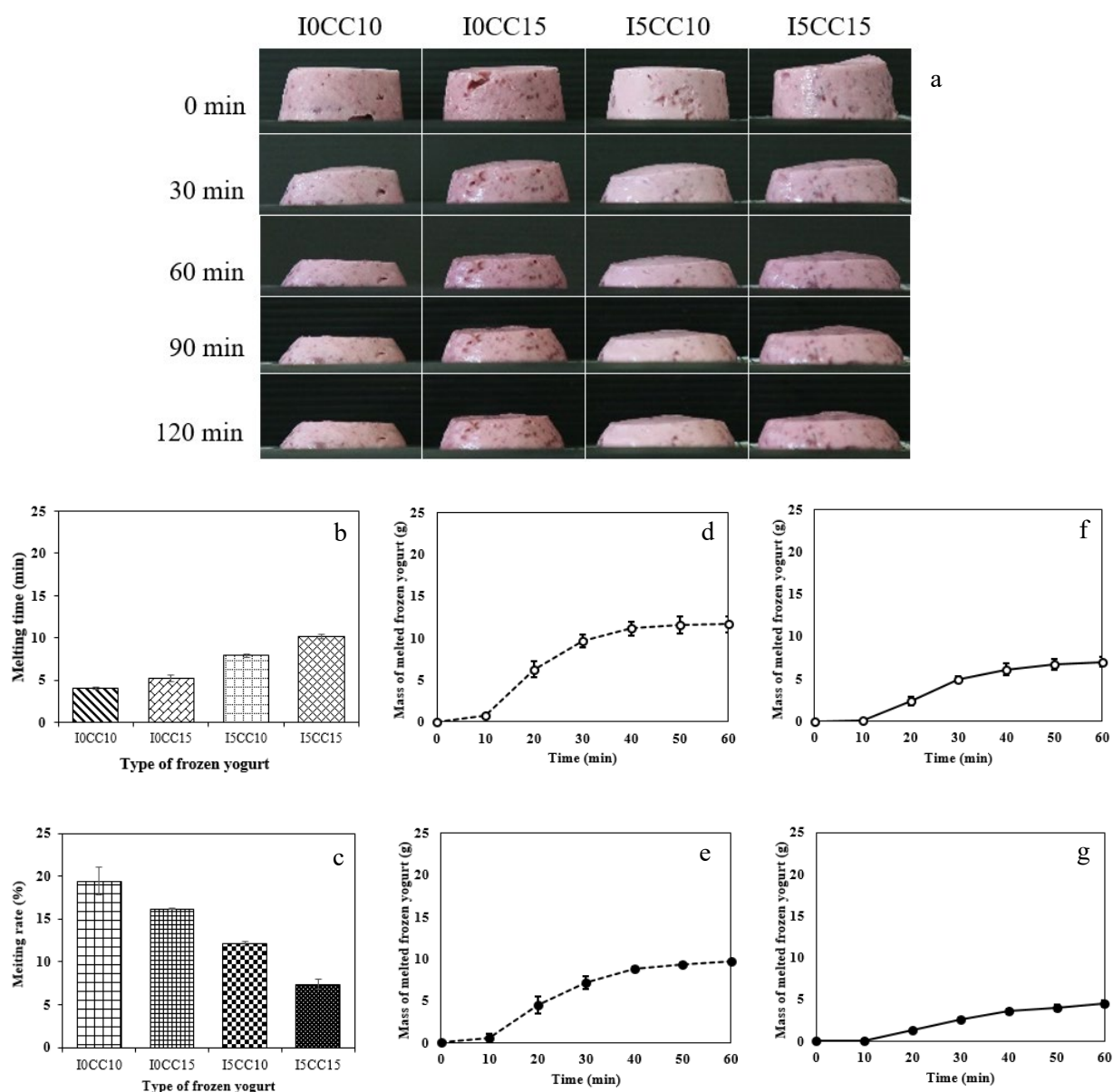
Figure 2. Melting profiles of frozen yogurts from different supplementation of inulin and CC contents; a) pictures taken during melting tests, b) the dripping of the first drop, c) melting rate and d) melted mass of frozen yogurts d) inulin content 0% and CC berry content 10%; I0CC10, e) inulin content 0% and CC berry content 15%; I0CC15, f) inulin content 5% and CC berry content 10%; I5CC10 and g) inulin content 5% and CC berry content 15%; I5CC15

3.3 Microbiological enumeration

Surviability of cultures is an important quality for frozen yogurt. Initial counts of lactic acid bacteria in the frozen yogurts from different supplementation of inulin and CC ranged between 6.29-6.72 log CFU/g. According to the sensorial evaluation, the sample with inulin 5% and CC 15% was monitored in order to study survivability of lactic acid bacteria in the frozen yogurt. Microbiological results revealed that LAB survival remained in the range of 6.58 to 5.90 log CFU/ g during 90 day-storage at -18 °C. LAB counts declined 0.68 log CFU/g compared to the initial counts. In the present study, only a slight reduction of LAB count was observed, although stress factors, e.g. sublethal injuries caused by freezing, the presence of oxygen in the product could affect the cell viability (Leando et al., 2013; Rezaei et al., 2014). LAB survival in this study could possibly indicate resistant to severe conditions of starter cultures and inulin supplementation could improve their survivability during freezing. These results are in agreement with previous reports by Muzammil et al. (2017), Hong and Marshall (2001) and Rezaei et al. (2014). The finding in this study confirmed that LAB in the frozen yogurt could remain viable in the CC frozen yogurt product and during frozen storage.

a

b
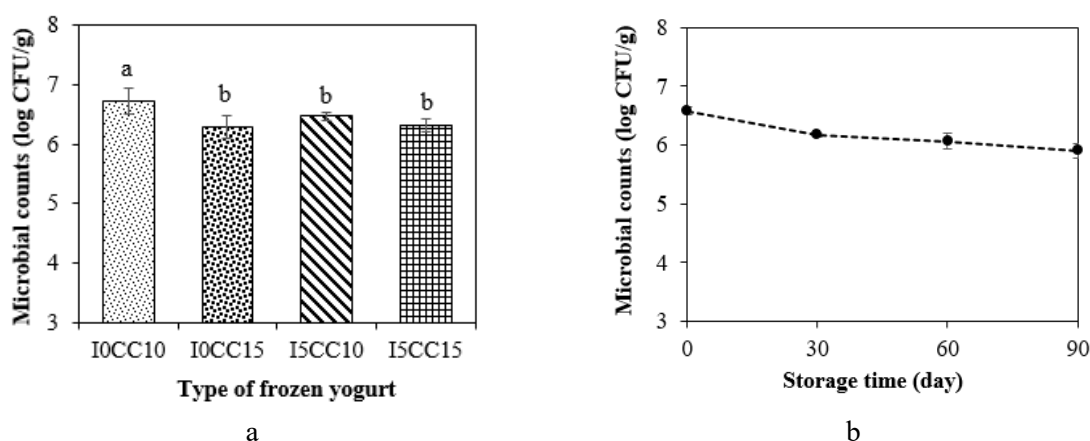
Figure 3. Initial viable counts of lactic acid bacteria in CC frozen yogurts from four treatments (a) and changes of viable counts in the frozen yogurt containing 5% inulin and 15% CC during storage at -18 °C (b), inulin content 0% and CC berry content 10%; I0CC10, inulin content 0% and CC berry content 15%; I0CC15, inulin content 5% and CC berry content 10%; I5CC10, and inulin content 5% and CC berry content 15%; I5CC15

3.4 Sensory analysis

Sensorial properties revealed the significant effect of supplementation of inulin and CC on the attributes of flavor, texture and overall preference in frozen yogurts ($p \leq 0.05$). Hedonic scores of a texture attribute was significantly higher in the frozen yogurts containing higher levels of inulin contents compared with the frozen yogurts containing lower levels of inulin contents (Figure 4). Inulin supplementation in the frozen yogurts considerably improved the texture attribute of CC frozen yogurts. These results are in agreement with melting properties of the frozen yogurts as previously discussed. There is no significant difference in overall preference scores between the frozen yogurts with and without inulin supplementation, whereas CC supplementation was the factor statistically influencing the overall preference scores of the frozen yogurts. Frozen yogurts containing 15% CC attained higher hedonic scores in the sensorial attribute of overall preference compared with frozen yogurts containing 10% CC ($p \leq 0.05$).
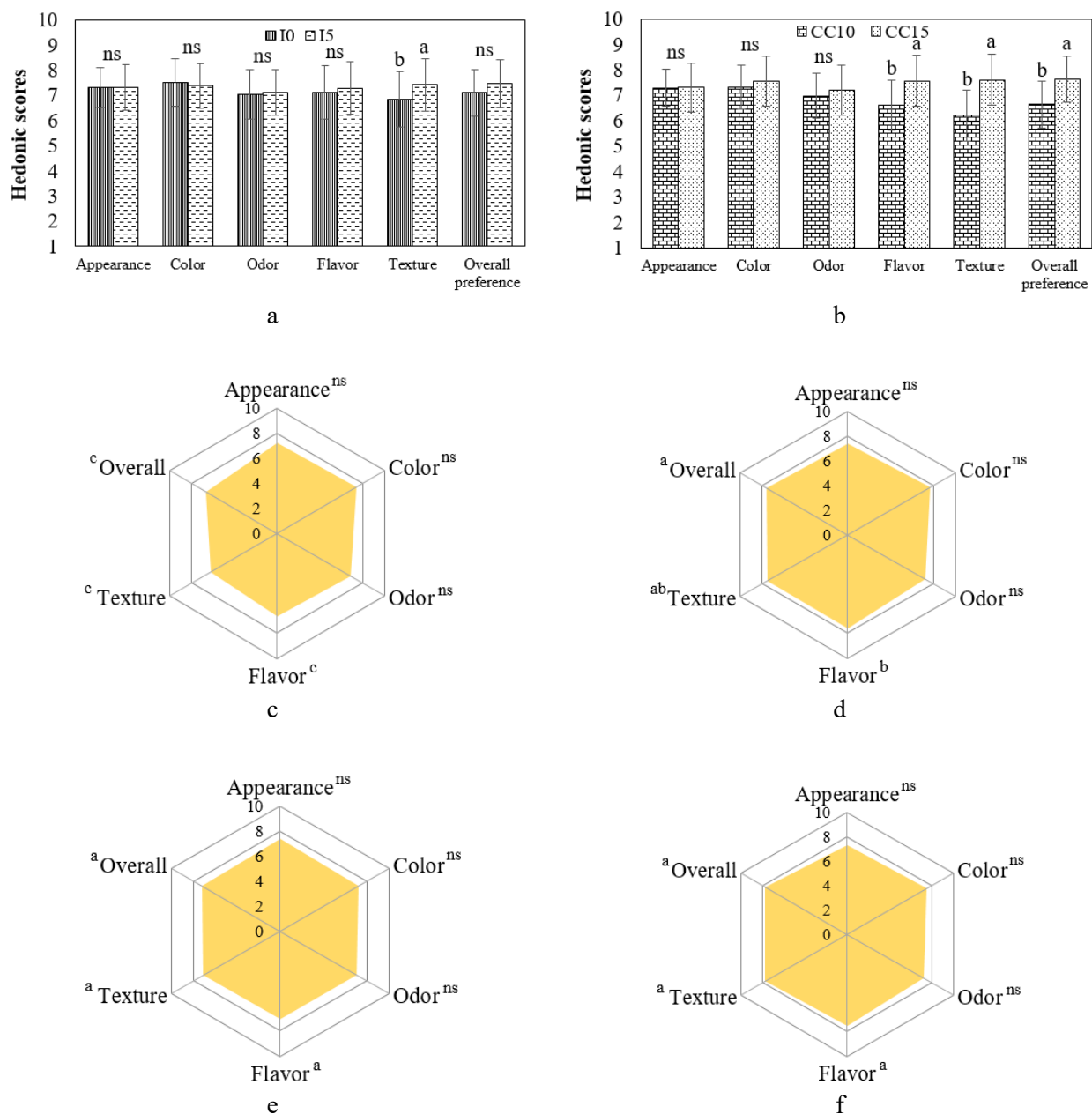
Figure 4. Effect of inulin a) and CC b) supplementation on sensory quality of frozen yogurts, c) inulin content 0% and CC berry content 10%, d) inulin content 0% and CC berry content 15%, e) inulin content 5% and CC berry content 10% and f) inulin content 5% and CC berry content 15%
ns denotes means are not significantly different (p>0.05) and means with different letters denote significantly different (p≤0.05).

4. CONCLUSION

This study suggests that inulin supplementation could prolong survival of lactic acid bacteria in CC frozen yogurt. Furthermore, inulin addition into the frozen yogurt mixture could elevate the melting times of the CC frozen yogurt. The conditions to achieve the appropriate production of frozen yogurt were supplementation of inulin and CC into the yogurt at the levels of 5 and 15%, respectively. Inulin and CC fruits have a potential to serve as frozen yogurt ingredients for a healthy diet delivering

multifunctional bioactive components, e.g. peptides, phytochemicals, prebiotics and live microorganisms.

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2:15 PM - 2:30 PM (Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room C)

[4-1330-C-04] Utilization of Banana Agricultural Waste: Effects of Processing Conditions on Properties of Unripe Banana (*Musa Cavendish*) Pulp and Peel Flours

*Natthawuddhi Donlao[1,2], Asia Perin[1], Nasuha Bunyameen[1] (1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai, Thailand(Thailand), 2. Innovative Food Packaging and Biomaterials Unit (IFP), Mae Fah Luang University, Thailand(Thailand))
Keywords: Antioxidant capacity, Banana peel flour, Banana pulp flour, Citric acid, Drying temperature, Resistant starch

Banana is one of the most important tropical fruits. Flour from green bananas contains a high amount of resistant starch that can reduce constipation, prevent colon cancer and provide benefit to the digestive system. Banana peel is an underused by-product which is known as a potential source of high dietary fiber, protein, essential amino acids, and total phenolic compounds. The objective of this study was to investigate the effects of citric acid pretreatments and drying temperatures on physical, chemical, and functional properties of unripe banana pulp and peel flours. Rejected green bananas (*Musa cavendish*) at the 2nd stage of maturity were obtained from Phaya Meng Rai Agricultural Limited Partnership in Phaya Meng Rai District, Chiang Rai, Thailand. Unripe bananas were washed with clean water and drained. Banana pulps and peels were then separated. The banana pulps and peels were pretreated with citric acid solution at different concentrations (i.e., 0.5, 0.7, and 1.0% w/v) and dried at different temperatures (i.e., 40, 60, and 80 °C). Moisture content, color, water holding capacity, and pasting properties of two flours were determined. Resistant starch of banana pulp flour and total polyphenol content and antioxidant activity of banana peel flour were also determined. In banana pulp flour, higher citric acid concentration and drying temperature resulted in a lower level of moisture content and water holding capacity. The resistant starch increased with increasing drying temperature. Highest peak viscosity was found at the condition of 0.7% citric acid pretreatment. In banana peel flours, total polyphenol content increased with increasing drying temperature. A higher concentration of the citric acid solution and lower drying temperature resulted in increasing *L** value. The highest value of antioxidant capacity was found in the sample dried at 60 °C. It is recommended that pretreating with 1.0% citric acid solution and drying at 80 °C can produce good quality of banana pulp flour. However, good quality of banana peel flour can be achieved by pretreating with 1.0% citric acid solution, followed by drying at 60 °C.

Oral Session | Food Quality

[4-1330-D] Food Quality (2)

Chair:Nurheni Sri Palupi(Bogor Agricultural University, Indonesia)

Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room D (4th room)

[4-1330-D-01] **Reducing Allergenicity of Soy Protein Isolate from Local Varieties of Soybean through Glycation with Lactose**

*Nurheni Sri PALUPI[1,2], Endang Prangdimurti[1,2], Didah Nur Faridah[1,2], Muhammad Hasriandy Asyhari[3]　(1. Department of Food Science and Technology, Faculty of Agricultural Engineering and Technology, Bogor Agricultural University(Indonesia), 2. Southeast Asian Food and Agricultural Science and Technology (SEAFAST) Center, Bogor Agricultural University(Indonesia), 3. Food Science Graduate Program, Graduate School, Bogor Agricultural University(Indonesia))

　1:30 PM - 1:45 PM

[4-1330-D-02] **Nutritional Quality of Fertilized and Salted Philippine Mallard Duck (** *Anas platyrhynchos L.***) Eggs Consumed in Victoria, Laguna, Philippines**

*Lotis Escobin Mopera[1], Pauline Saludo[1], Floirendo Flores[1], Ma. Josie Sumague[1], Bryan Rey Oliveros[1], Wilson Tan[1]　(1. Institute of Food Science and Technology, University of the Philippines Los Banos(Philippines))

　1:45 PM - 2:00 PM

[4-1330-D-03] **Efficacy of 1-methylcyclopropene (1-MCP) Post-cutting Treatment on the Storage Quality of Fresh-cut 'Queen' Pineapple (Ananas comosus(L.) Merr. cv. 'Queen')**

*Meryl Ancheta Bernardino[1], Katherine Anne Castillo Israel[1], Edralina Serrano[1], James Bryan Gandia[1], Wella Absulio[1]　(1. University of the Philippines Los Banos(Philippines))

　2:00 PM - 2:15 PM

[4-1330-D-04] **Effect of Direct and Indirect Heating On Heat Stability of Goat Milk**

*Souvia Rahimah[1]　(1. Universitas Padjadjaran(Indonesia))

　2:15 PM - 2:30 PM

Oral Session | Food Quality
[4-1330-D] Food Quality (2)
Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room D (4th room)

[4-1330-D-01] Reducing Allergenicity of Soy Protein Isolate from Local Varieties of Soybean through Glycation with Lactose

*Nurheni Sri PALUPI[1,2], Endang Prangdimurti[1,2], Didah Nur Faridah[1,2], Muhammad Hasriandy Asyhari[3] （1. Department of Food Science and Technology, Faculty of Agricultural Engineering and Technology, Bogor Agricultural University(Indonesia), 2. Southeast Asian Food and Agricultural Science and Technology (SEAFAST) Center, Bogor Agricultural University(Indonesia), 3. Food Science Graduate Program, Graduate School, Bogor Agricultural University(Indonesia)）

Keywords: Allergenicity, Enzyme-Linked Immunosorbent Assay (ELISA), Glycation degree, Soy protein isolate (SPI), SPI-lactose conjugate

Food allergy is a specific immunological response caused by allergens contained in food. Soybean is one of the eight type of food products that often cause allergies that can reduce its quality in term of safety aspect. The soybean processing can reduce the risk of allergies by modifying the structure of the soy protein so as to produce hypoallergenic food. Processing involving the Maillard reaction by conjugating proteins with reducing sugars has the potential to reduce allergenicity. This research aims to: (1) determine the degree of glycation based on the formation of soy protein isolates (SPI)-lactose conjugate and free amino acid; (2) determine the molecular weight profile of SPI and SPI-lactose conjugate: and (3) analyze the allergenicity of SPI-lactose conjugate compared to SPI. Protein isolation was carried out by precipitation of soybean protein of Anjasmoro and Grobogan varieties at their isoelectric point by pH arrangement. Then the SPI was reacted with lactose under pH 9.5 and 95°C for 60 minutes. Determination of glycation degree of SPI-lactose conjugate was carried out using two methods, namely thiobarbituric acid reactive substances (TBARS) method and Bradford method for free amino acid. The protein molecular weight profile was analyzed using the SDS-PAGE electrophoresis method. Alergenicity of SPI and SPI-lactose conjugate were analyzed quantitatively using the Enzyme-Linked Immunosorbent Assay (ELISA) method. The results showed that the higher protein content in SPI cause higher glycation degree of the SPI-lactose conjugate. SDS-PAGE electrophoresis results showed that the molecular weight profiles of Anjasmoro and Grobogan SPI were 11.3-144.2 kDa and 10.7-159.0 kDa. The glycation process can eliminate or reduce the intensity of protein bands that are suspected to be the major allergen proteins in soybeans, namely Gly m Bd 60K, Gly m Bd 30K or P34, and Gly m Bd 28K. The glycation reaction can reduce allergenicity in local varieties soybeans (Anjasmoro and Grobogan) by 43.12% and 29.85%.

Reducing Allergenicity of Soy Protein Isolate from Local Varieties of Soybean through Glycation with Lactose

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ABSTRACT

Food allergy is a specific immunological response caused by allergens contained in food. Soybean is one of the eight type of food products that often cause allergies that can reduce its quality in term of safety aspect. The soybean processing can reduce the risk of allergies by modifying the structure of the soy protein so as to produce hypoallergenic food. Processing involving the Maillard reaction by conjugating proteins with reducing sugars has the potential to reduce allergenicity. This research aims to: (1) determine the degree of glycation based on the formation of soy protein isolates (SPI)-lactose conjugate and free amino acid; (2) determine the molecular weight profile of SPI and SPI-lactose conjugate: and (3) analyze the allergenicity of SPI-lactose conjugate compared to SPI. Protein isolation was carried out by precipitation of soybean protein of Anjasmoro and Grobogan varieties at their isoelectric point by pH arrangement. Then the SPI was reacted with lactose under pH 9.5 and 95°C for 60 minutes. Determination of glycation degree of SPI-lactose conjugate was carried out using two methods, namely thiobarbituric acid reactive substances (TBARS) method and Bradford method for free amino acid. The protein molecular weight profile was analyzed using the SDS-PAGE electrophoresis method. Alergenicity of SPI and SPI-lactose conjugate were analyzed quantitatively using the Enzyme-Linked Immunosorbent Assay (ELISA) method. The results showed that the higher protein content in SPI cause higher glycation degree of the SPI-lactose conjugate. SDS-PAGE electrophoresis results showed that the molecular weight profiles of Anjasmoro and Grobogan SPI were 11.3-144.2 kDa and 10.7-159.0 kDa. The glycation process can eliminate or reduce the intensity of protein bands that are suspected to be the major allergen proteins in soybeans, namely Gly m Bd 60K, Gly m Bd 30K or P34, and Gly m Bd 28K. The glycation reaction can reduce allergenicity in local varieties soybeans (Anjasmoro and Grobogan) by 43.12% and 29.85%.

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Oral Session | Food Quality
[4-1330-D] Food Quality (2)
Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room D (4th room)

[4-1330-D-02] Nutritional Quality of Fertilized and Salted Philippine Mallard Duck (*Anas platyrhynchos L.*) Eggs Consumed in Victoria, Laguna, Philippines

*Lotis Escobin Mopera[1], Pauline Saludo[1], Floirendo Flores[1], Ma. Josie Sumague[1], Bryan Rey Oliveros[1], Wilson Tan[1] （1. Institute of Food Science and Technology, University of the Philippines Los Banos(Philippines)）
Keywords: Philippine Mallard Duck, Fertilized Duck Eggs, Salted Duck Eggs, Nutritional Quality

Duck industry is considered as the second largest in the Philippine poultry industry that contributes to farmer's income through egg and meat production. Ducks are mainly raised for the production of boiled fertilized eggs known locally as *balut* and salted duck eggs.The recent increase in the demand and consumption of these commodities because of diet diversification and utilization, fostered the growth in the duck industry as well as assure sustainable supply of the raw materials for utilization in processing *balut* and salted eggs. To help establish standards for both the quality and safety of *balut* and salted egg, the nutritional property of these two commodities was evaluated in one of the popular towns known to produce *balut* and salted duck eggs. Proximate, mineral and fatty acid analyses of *balut* and salted eggs were done. Both *balut* and salted duck eggs were found to contain considerable amount of protein and calories at 11% and 100%, respectively. In general, the major fatty acids found in duck eggs are oleic acid (C18:1), myristic acid (C14:0) and linolenic acid (C18:2). Salted eggs contained more oleic acid (52.18%) while *balut* has more myristic acid (26.30%). Salted eggs showed higher sodium content as affected by the clay-salt curing process. However, the level of salt still conforms within the recommended nutrient intake for sodium. This study contributes in addressing research gaps in the lack of information for standards and marketing distribution of these products as well as provide information on the nutritional comparison with similar commodities.

Nutritional Quality of Fertilized and Salted Philippine Mallard Duck (*Anas platyrhynchos L.*) Eggs Consumed in Victoria, Laguna, Philippines

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ABSTRACT

Duck industry is considered as the second largest in the Philippine poultry industry that contributes to farmer's income through egg and meat production. Ducks are mainly raised for the production of boiled fertilized eggs known locally as *balut* and salted duck eggs. The recent increase in the demand and consumption of these commodities because of diet diversification and utilization, fostered the growth in the duck industry as well as assure sustainable supply of the raw materials for utilization in processing *balut* and salted eggs. To help establish standards for both the quality and safety of *balut* and salted egg, the nutritional property of these two commodities was evaluated in one of the popular towns known to produce *balut* and salted duck eggs. Proximate, mineral and fatty acid analyses of *balut* and salted eggs were done. Both *balut* and salted duck eggs were found to contain considerable amount of protein and calories at 11% and 100%, respectively. In general, the major fatty acids found in duck eggs are oleic acid (C18:1), myristic acid (C14:0) and linolenic acid (C18:2). Salted eggs contained more oleic acid (52.18%) while *balut* has more myristic acid (26.30%). Salted eggs showed higher sodium content as affected by the clay-salt curing process. However, the level of salt still conforms within the recommended nutrient intake for sodium. This study contributes in addressing research gaps in the lack of information for standards and marketing distribution of these products as well as provide information on the nutritional comparison with similar commodities.

Keywords: Philippine Mallard Ducks, Fertilized Duck Eggs, Salted Eggs, Nutritional Quality

1. INTRODUCTION

Processing and consumption of duck eggs has long been done in Asian countries particularly in China, South Korea, Bangladesh, Thailand, Vietnam, Lao, Malaysia, Singapore and Philippines (Tang et al., 2019; Ahmad et al., 2017; Ganesan et al., 2014; Chang and Dagaas, 2004). Apart from its importance as an integral part of the food culture in these countries, duck eggs were also reported as a good source of protein and other nutrients and is regarded as a food with high nutritional quality (Ahmad et al., 2017; Al-Obaidi and Al-Shadeedi, 2016). People eat duck eggs for its high nutritional value because of the optimal composition of essential amino acids and the considerable composition of fatty acids with a high percentage of polyunsaturated fatty acids and a favorable ratio of omega 6- to omega 3-fatty acids. In addition, it is economical as well as quick and easy to prepare and serve.

In the Philippines, the duck industry contributes to 20.32% of the 55.4 billion peso contribution of the poultry industry in the country's Gross Domestic Product (GDP) (PSA, 2019). The duck egg volume of production reached a total of 46.61 thousand metric tons in 2019 (PSA, 2019). This volume grew by 2.60 percent relative to its previous year's level of 45.43 thousand metric tons (PSA, 2019). In general, about 87% of the total duck egg production is processed into *balut* (fertilized duck egg embryo), 7% to salted eggs and the remaining 6% for other duck egg products like century eggs and *penoy* (Chang & Dagaas, 2004).

In terms of production and processing of duck eggs, the town of Victoria in the province of Laguna was dubbed as the "Duck Raising Center of the Philippines" (Atienza et al., 2015). According to the Department of Agriculture (2003), the duck industry in Victoria is a 5.5 billion peso industry which contributes a total duck egg production of 2.5 billion pesos. The town is known for its duck products which include meat and eggs, particularly *balut* and salted egg.

Balut is produced by incubating fertilized duck egg at 40-42.5°C with high humidity. After 18 days, a partially developed embryo can be seen during candling. These "embryonated eggs" are harvested, boiled and sold as *balut*. *Balut* is known to be good but inexpensive source of protein and calcium (Magat, 2002)

along with other nutrients as stated in the Philippine Composition Table (FNRI, 1997). In practice, the infertile eggs can either be processed to salted egg, century egg or *penoy*.

Salted egg is produced by brining or by curing in clay. In most duck farms in the Philippines, curing in clay is often used. This method involves coating duck eggs with a mixture of clay and salt then stored indoors at room temperature for 18 days. After curing, salted eggs are boiled for 20-30 mins prior to consumption. Some producers cover the egg shell with red food colorings or markings for salted eggs to differentiate them from fresh eggs and other egg products as part of the marketing strategy for this egg product.

Both *balut* and salted duck eggs are consumed based on the fact that eggs, in general, are good sources of protein and fat. It also contains dietary macro-minerals (Ca, P, Na, K, Mg) and trace minerals (Fe, Zn, Cu, Mn) (FNRI, 1997). Duck eggs, as in the case of *balut* and salted eggs, are preferred over hen eggs because of its larger size (about 30% bigger) and higher nutritional value which is attributed to higher fat content found in duck eggs (Ahmad et al., 2017). This is because duck eggs contain relatively less water and higher percentage of proteins and fats in the yolk, albumen and total contents of egg as compared to chicken eggs (Rahman et al., 2010).

Variability in the nutritional composition of eggs are dependent on several factors. More often, it is affected by the kind of nutrients fed to the animal. In the Philippines, the feeds for the ducks are normally supplemented with other agricultural products found within the vicinity or location of the duck farms. Another factor that affects the quality of nutrients in duck eggs is the manner of processing. Processing can alter the nutritional composition of the eggs (Ganesan et al., 2014). Thus, it is important to maintain the nutritional quality of the duck eggs even after processing. The maintenance of the egg quality from the time of their production till their delivery to the final consumer is of great importance (Rahman et al., 2010). Information on nutritional composition of duck specifically from the town of Victoria can be a valuable information especially on the marketing strategy of the community involved in processing of the duck eggs from the duck farms in this area. Apart from its economic value, it is necessary to know the nutritional quality of duck eggs for consumer's satisfaction. In addition, the data that was generated from this study on

the nutritional quality can be a benchmark for other localities engaged in duck production and processing, particularly, *balut* and salted duck eggs in the Philippines as well as other countries in Asia engaged in duck production and processing. Thus, a comparative study on the total nutritional quality and value of fresh, fertilized and salted duck eggs from Victoria, Laguna, Philippines were explored.

2. MATERIALS AND METHODS

2.1 Egg Samples

Convenience sampling was employed in the selection of egg samples from outlet stores in Victoria, Laguna from May to July 2018. *Balut* eggs were immediately boiled for 45 mins. All eggs samples were carefully cracked, shells removed and homogenized in the Osterizer blender prior to analysis. Additional sample preparation were done in accordance with succeeding analyses.

2.2 Determination of Proximate and Nutrient Composition of Duck Eggs

Proximate Analysis. Proximate analysis such as moisture, protein, fat, ash and carbohydrate were determined according to AOAC method (2000). Moisture content was determined by oven drying at 105 $\pm 5^0$C. Protein content (x 6.25) was determined by Kjeldahl method as stated in AOAC (2000). The fat content was determined by Soxhlet method. Ash was determined by incineration of the dried sample at 600^0C for 5 h (AOAC, 2000). The carbohydrate content was computed as nitrogen free extract (NFE) by subtracting the moisture, protein, fat and ash content**.**

2.3 Mineral Analysis. Egg samples were hydrolyzed by wet ashing (AOAC, 2000) using strong acids: nitric acid (HNO_3), perchloric acid ($HClO_4$) and hydrochloric acid (HCl) prior to mineral analysis. Atomic absorption spectrophotometry was used to quantify the minerals in all samples. All mineral concentrations were reported in parts per million (ppm).

2.4 Fatty Acid Profile of Oils extracted from Duck Eggs. Petroleum ether was used to extract oils from egg samples by shaking for 1 h. The extracted oils were placed in vials for storage in the freezer until analysis. Oil samples were submitted to the Central Analytical Services Laboratory, National Institute

for Molecular Biology and Biotechnology, (BIOTECH-UPLB) for fatty acid profiling against fatty acid methyl ester standards using Gas Chromatograph (AOAC 969.33; 963.22, 2000).

2.5 Statistical Analysis

All of the experiments were performed in triplicates, and the results are expressed as means ± SD. Statistical analyses were performed using the Student's t-test. Differences were considered significant at P < 0.05. Means are compared using T-test at P <0.05%.

3. RESULTS AND DISCUSSION

3.1 Proximate Composition of Fresh Duck Eggs, *Balut* and Salted Eggs

Nutritional quality is often associated with the amount of basic nutrients found in certain food products. Carbohydrate, fat and protein content in most food are used as indices whether certain food items are important sources of those particular nutrients. In general, eggs are known for its protein, fat and mineral content. However, it also contains other nutrients such as carbohydrate, free amino acids and vitamins.

Proximate composition of duck eggs are shown in Table 1. Ash content in food materials is often use as an index of the mineral content. Previous studies (Ahmad et al. 2017; Zhao et al., 2014; Ganesan et al., 2014) have reported ash content of duck eggs in both fresh and salted eggs. The amount of ash obtained ranged from 1.01-1.87%, these values were comparable to the ash content from duck eggs as reported by Ahmad et al. (2017).

Moisture content of *balut* (65.95 ± 0.06%) was significantly lower compared to fresh duck egg (72.19 ± 0.32%) and salted egg (72.74 ± 0.83). Moisture loss in *balut* can be attributed to the longer processing time (45mins) compared to salted eggs (25mins). Processing lowers the moisture content of duck eggs, a phenomenon, also observed in other food and agricultural products. Further, prolonged heat treatment causes slight shrinking of the contents of eggs. *Balut* is also found to contain higher amount of fiber (4.33 ± 0.28%) due to the partially developed feathers of the embryo. The presence of the

underdeveloped duck embryo is characteristic to fertile duck eggs incubated for about 16-18 days or even 20 days in other countries.

Processing alters the protein composition. This was observed in the amount of protein before and after processing of the duck eggs. In this study, duck eggs were found to be an extensive source of protein but the protein content of both *balut* and salted eggs used in this study decreased during processing. The values obtained ranged from 11.49 to 15.54 %, the latter was exhibited by the fresh duck eggs. These values were comparable to that reported by Dirwan-Muchlis and Nurcholis (2019). At the cellular level, high temperature increases the kinetic energy of protein molecules that leads to their denaturation and later to the formation of stronger covalent bonds with other protein molecules. The previously attached water molecules to the proteins are now released resulting in moisture loss and hardened egg contents. From $15.54 \% \pm 3.73$ proteins in fresh eggs, only $11.89 \pm 1.28\%$ and $11.49 \pm 2.29\%$ for *balut* and salted eggs, respectively, were retained after processing. During protein degradation, proteins are broken down to their primary structures in the form of amino acids. Some amino acids form volatile compounds under alkali conditions which might have also transferred to the curing solution (Zhao et al., 2014), as in the case of salted eggs.

On the other hand, higher amounts of lipids are observed in salted eggs ($10.15 \pm 0.11\%$). Lipids in eggs exist as low-density lipoproteins (LDL) in the yolk plasma (Gilbert, 1971). During clay-curing, sodium chloride (NaCl) leads to dehydration and destruction of LDL structures. Some of the lipids of the cooked yolk become free (Lai et al., 1997) and contributed to the total fat obtained as shown in Table 1.

However, contrary to the results presented by Wang et al. (2014) that the fat content in eggs can be reduced to 0.61% by salting, this study showed that the fat content of the salted duck eggs had a significant increase in fat content compared to fresh duck egg and *balut*.

Table 1. Proximate composition of fresh, *balut,* and salted duck eggs.

Composition*, %	Fresh Egg	*Balut*	Salted Egg
Moisture	72.19 ± 0.32^a	65.95 ± 0.06^b	72.74 ± 0.83^a
Fat	4.49 ± 0.47^b	4.01 ± 0.19^b	10.15 ± 0.11^a
Crude Fiber	2.42 ± 0.19	4.33 ± 0.28^a	1.07 ± 1.11
Protein	15.54 ± 3.73^a	11.89 ± 1.28^b	11.49 ± 2.29^b
Ash	1.28 ± 0.13	1.63 ± 0.42	1.01 ± 0.13

*Means with the same superscript are not significantly different from each other at $P < 0.05$.

3.2 Mineral Composition of *Balut* and Salted Eggs

Table 2 shows the amount of trace minerals of fresh, *balut* or salted egg. Results indicated that processing into *balut* and salted egg does not significantly affect the amount of minerals found in duck eggs. In particular, magnesium (Mg), zinc (Zn), copper (Cu) and manganese (Mn), ranged from 0.16-0.24mg/100g, 0.04-0.06mg/100g, 0.01-0.11mg/100g and 2-4µg/100g, respectively. Results, further exhibited that iron (Fe), was present at higher concentration in *balut* (0.51mg/100g) compared to fresh duck and salted eggs. The iron content however, is lower than the amount reported by Ganesan et al. (2014). Potassium is mostly lost during cooking, as manifested by its low concentration in *balut* (1.85mg/100g). In the case of salted eggs, curing played an important role in the concentration of inorganic minerals. Significant amounts of K (4.57mg/100g), Na (13.28mg/100g), and Ca (4.07mg/100g) were observed for salted eggs as shown in Table 2. The clay-salt mixture provided a barrier to minimize mineral losses in duck eggs while allowing minerals from the mixture to penetrate the egg shell and migrate into the eggs. In general, the mineral content of the duck eggs used in this study were ten times lower compared to previous studies reported (Durwan Muchalis and Nuchalis, 2019; Ahmad et al., 2019; Ganesan et al., 2014).

Table 2. Mineral composition of duck eggs.

Minerals (mg/100g)*	Fresh Eggs	*Balut*	Salted Eggs
P	4.44 ± 0.36 [a]	4.92 ± 0.11 [a]	3.76 ± 0.01 [b]
K	3.92 ± 0.02 [b]	1.85 ± 0.02 [c]	4.57 ± 0.01 [a]
Na	5.04 ± 0.01 [b]	1.24 ± 0.05 [c]	13.28 ± 0.02 [a]
Ca	3.61 ± 0.00 [b]	3.47 ± 0.02 [b]	4.07 ± 0.01 [a]
Fe	0.14 ± 0.01 [b]	0.51± 0.02 [a]	0.10 ± 0.01 [b]
Mg	0.29 ± 0.01 [a]	0.16 ± 0.01 [a]	0.24 ± 0.01 [a]
Mn	0.002 ± 0.0006 [a]	-	0.004 [a]
Zn	0.04 [a]	0.06 [a]	0.04 [a]
Cu	0.01 [b]	0.01 [b]	0.11 ± 0.01[a]

*Means with the same superscript are not significantly different from each other at P <0.05.

3.3 Nutritional Value of *Balut* and Salted Eggs

Duck eggs weigh about 65 g on the average and provide 79-91 kcal based from the results presented in Table 3. Protein was found to provide 7-10g/65g or 4-10% of the recommended dietary allowance for adults (FNRI, 1997). This value means that duck eggs are considered as a good source of protein.

Additional recommendation of the Food and Nutrition Research institute (PDRI, 2015) for adult male 19-39 years of age, is to limit the sodium intake to <2 g and increase the intake of potassium to about 3, 510 mg in adult. Table 3 shows that salted eggs exhibited the highest sodium intake per serving which is estimated at 863 mg/65 g. Potassium, on the other hand, was computed at 297 mg/ 65 g of duck eggs. High salt content for salted egg is as expected because of the processing method used to produce these products. In related studies, salt content in salted eggs in general, ranged from 7-10% after curing for 15- 30 days (Wang, 2017).

However, the estimated cholesterol content of salted eggs is 202.00 mg as calculated based from a study conducted by Aziz et al. (2012). The cholesterol content of duck egg with 60 g average weight yolk

proportion is 186.46 mg. In the Nutritional Guidelines for the Prevention of Heart Diseases and Diabetes Mellitus (FNRI-DOST, 2002), it was stated that the dietary cholesterol should be less than 300 mg/day. With the estimated cholesterol content, duck egg already provides 67% of the recommended dietary cholesterol.

Therefore, inclusion of duck eggs, particularly salted eggs, should be done in moderation due to its big contribution to dietary cholesterol and minimal contribution to mineral intake.

Table 3. Composition (per serving size of 65g) of fresh and processed duck eggs.

Components	Fresh Eggs	*Balut*	Salted Eggs
Total Fat, g	3	3	7
Total Carbohydrates, g	3	8	2
Fiber, g	2	3	0.6
Protein, g	10	8	7
Calories, kcal	80	90	100
Na, mg	3.3	0.8	8.6
K, mg	2.5	1.2	3.0

3.4 Fatty Acid Composition of *Balut* and Salted Eggs

The amount of fatty acids is affected by the nutrients incorporated in the feeds given to ducks. Duck eggs are found to contain more unsaturated fatty acids than saturated ones. The major fatty acids in egg lipids are oleic, myristic and linoleic acids found in duck eggs from Victoria, Laguna, Philippines are shown in Table 4. Salted eggs had the highest amount of oleic acid (51.57%); *balut* has more myristic acid (26.30%). Linoleic acid is found to decrease after processing from 12.52% in fresh eggs to 10% in processed eggs (10.34% in *balut* and 10.04% in salted eggs). This is in contrast to that reported by Men et al. (2015)

wherein palmitoleic acid and linoleic acid took a greater proportion in the unsaturated free fatty acids, and their contents increased during the pickling period of salted duck eggs.

Myristic and lauric acid are strongly correlated with higher cholesterol levels (German & Dillard, 2010). Combined myristic and lauric acid for duck eggs range from 27.28 – 29.34%. This should be considered in the consumption and inclusion of duck eggs in the diet.

Table 4. Fatty acids in duck eggs as compared to coconut oil.

Fatty acid (% distribution)	Fresh Egg	Salted egg	*Balut*
C10:0	0.58	0.46	0.69
C12:0	2.29	4.24	1.58
C14:0	26.27	25.10	26.20
C16:0	2.15	1.31	0.59
C18:0	2.88	ND	3.62
C18:1	46.93	52.18	51.57
C18:2	12.52	10.34	10.04

4. CONCLUSION

Processing into salted eggs and *balut* has significantly affected the nutritional value of fresh duck eggs, including but not limited to their proximate composition and nutritional contents. Proteins, compared to other macronutrients, underwent the most biochemical and structural changes during processing. *Balut* and salted eggs provide more energy than fresh duck eggs based on the results of this study. Curing of eggs in clay-salt does not increase sodium levels in the salted eggs that could pose a health issues, however, consumption of these eggs must be done in moderation since they contain high levels of cholesterol.

ACKNOWLEDGMENTS

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2:00 PM - 2:15 PM (Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room D)

[4-1330-D-03] Efficacy of 1-methylcyclopropene (1-MCP) Post-cutting Treatment on the Storage Quality of Fresh-cut 'Queen' Pineapple (Ananas comosus(L.) Merr. cv. 'Queen')

*Meryl Ancheta Bernardino[1], Katherine Anne Castillo Israel[1], Edralina Serrano[1], James Bryan Gandia[1], Wella Absulio[1] (1. University of the Philippines Los Banos(Philippines))
Keywords: visual quality rating, 1-MCP, ethylene suppresant, fresh-cuts, Queen pineapple

The efficacy of 1-methylcyclopropene (1-MCP) applied as a post-cutting treatment on freshcut 'Queen' pineapple was determined in order to assess its potential to maintain the storage quality of fresh-cut 'Queen' pineapple, a major Philippine variety. 1-MCP at a concentration of 1 uL• L -1 was applied as post-cutting treatment by injecting the gas into packed fresh-cut 'Queen' pineapples in polypropylene tray overwrapped with LDPE stretchable film. The packed fruits were stored at 5 oC and monitored for headspace gas concentrations (ethylene, CO_2, O_2), visual quality deterioration parameters and microbial deterioration indicators. 1-MCP was found to effectively elicit its ethylene inhibiting action as shown by lowered headspace ethylene by about 40% at day 4 storage. Headspace CO_2 levels were likewise lowered by 1-MCP to about 50% at day 2 while higher headspace O_2 levels were generally obtained which had the highest increase at day 2 (about 18%) which created an improved modified atmosphere condition inside the package compared with the control. No significant effects on the visual quality were noted throughout storage. Color differences were however observed, with 1-MCP treatments having significantly higher lightness values and higher hue values at day 2. 1-MCP did not affect the microbial growth (aerobic bacteria, acid-forming bacteria, yeasts and molds, coliforms) on the samples during storage. Aerobic bacteria count was slightly lower than the control at day 3. The fresh-cut pineapple packaged in the manner described had a shelf-life of 3 days based on the microbial limits set by EU countries which is log 7 cfu/mL aerobic plate count. To the best of our knowledge, this is the first study which demonstrated the effects of 1-MCP on fresh-cut pineapple of the 'Queen' variety.

2:15 PM - 2:30 PM (Wed. Sep 4, 2019 1:30 PM - 2:30 PM Room D)

[4-1330-D-04] Effect of Direct and Indirect Heating On Heat Stability of Goat Milk

*Souvia Rahimah[1] (1. Universitas Padjadjaran(Indonesia))
Keywords: goat milk, direct heating, indirect heating, heat stability

Goat milk is known to be generally less heat stable than cow milk. Two different genotypes of goat skim milk: Dahlem Cashmere (DC) and Saanen (SA) and cow milk as a reference, were subjected to three standard heating procedures under pilot plant conditions, using direct (steam injection) and indirect (tubular heat exchanger) techniques. Direct steam injection carried out under UHT conditions (140 ^{o}C, 0 s) with and without preheating treatment. Indirect heating procedures included high temperature-short time (HTST) pasteurisation (90 ^{o}C), extended short life (ESL) (120 ^{o}C) and ultra high temperature (UHT) (140 ^{o}C) treatments for 0, 60 and 120 s. The heated milks were analyzed with regard to the degree of whey protein denaturation (HPLC), casein micelles size (dynamic light scattering) and calcium content in serum (EDTA titration).

Analysis results indicated no severe difference between tube and pilot plant trials at 140 ˚C for 0 s, except for casein micelle size, DC milk from pilot plant trials with preheating (90 ˚C for 120 s) contained bigger particles.

Oral Session | Postharvest Facility

[4-1445-C] Postharvest Facility

Chair:Ahmad Al-Mallahi(Dalhousie University, Canada)

Wed. Sep 4, 2019 2:45 PM - 3:30 PM Room C (3rd room)

[4-1445-C-01] The Effect of Level of Fill on Nutritional Quality of Maize in an Un-aerated Clay Silos

*Mobolaji Omobowale[1], Jonathan Ogwumike[1] （1. University of Ibadan(Nigeria)）

2:45 PM - 3:00 PM

[4-1445-C-02] Pod Storage and Maturity Effects on Specialty Cacao Pulp Quality

*Jeana Cadby[1], Tetsuya Araki[1], Ian Marc Cabugsa[2] （1. University of Tokyo, Dept. Global Agricultural Sciences(Japan), 2. Ateneo de Davao University, Dept. of Chemistry(Philippines)）

3:00 PM - 3:15 PM

[4-1445-C-03] Current Status of Monitoring Post-Harvest Potato Storage Units in Atlantic Canada

*Ahmad Al-Mallahi[1] （1. Dalhousie University(Canada)）

3:15 PM - 3:30 PM

2:45 PM - 3:00 PM (Wed. Sep 4, 2019 2:45 PM - 3:30 PM Room C)

[4-1445-C-01] The Effect of Level of Fill on Nutritional Quality of Maize in an Un-aerated Clay Silos

*Mobolaji Omobowale[1], Jonathan Ogwumike[1] (1. University of Ibadan(Nigeria))
Keywords: Clay Silo, Grain Quality, Postharvest losses, Maize, Level of Fill

Silos have a fixed volume at the time of construction; harvests however vary from season to season. Arising from complaints of severe postharvest losses in grains, most especially in the humid tropics of southern Nigeria, this study investigated the effect of level of filling on the nutritional quality of stored maize. Three clay silos labelled S_1 to S_3 of the same dimension and capacity (3.5 metric tonnes) were used. The walls were painted externally with gloss paint to eliminate or reduce water absorption into the silos. The three silos were filled with shelled maize; with the first silo (S_1) was filled with 3 tonnes, the second silo (S_2) was filled with 2 tonnes while the third silo (S_3) was filled with a tonne of maize. A storage experiment was carried out for 6 months and data was collected on environmental conditions within the silo as well as grain quality with increasing time of storage. Temperature, relative humidity and equilibrium moisture content were obtained three times daily for each of the three silos, focusing on the air-space between the grains and the head-space above the grains. Temperature fluctuations within the fully filled S_1 as indicated by the statistical range of 2°C was minimal compared with that of S_2 and S3 at 7 and 8°C respectively. Silo S_3 showed high values of relative humidity toward the end of storage period, ranging between 65% to 80% at the top level and 76% at the bottom level. Equilibrium moisture content at the airspace above the grains in S_1 increased from 12.0% at the beginning of storage in the dry season to 15.6% at the end of storage in the rainy season compared with S_2 and S_3 which increased from 12.0 to 17% and 12.0 to 17.2% respectively within the same period. Fungal and total viable counts showed an increase in microbial activity with decreasing level of fill. Grains were found to be more prone to deterioration with decreasing level of fill and recommendations were made to farmers and grain traders on proper actions to take as the situation arises.

3:00 PM - 3:15 PM (Wed. Sep 4, 2019 2:45 PM - 3:30 PM Room C)

[4-1445-C-02] Pod Storage and Maturity Effects on Specialty Cacao Pulp Quality

*Jeana Cadby[1], Tetsuya Araki[1], Ian Marc Cabugsa[2] (1. University of Tokyo, Dept. Global Agricultural Sciences(Japan), 2. Ateneo de Davao University, Dept. of Chemistry(Philippines))
Keywords: pod storage, post harvest quality, Theobroma cacao, specialty cacao

The quality of the cacao fruit pulp surrounding the seeds plays an important role in the early stages of post harvest processing, namely fermentation, influencing final cacao quality for specialty cacao products. Pod storage is common in cacao production, and has also been cited as a technique used by bulk/commodity cacao producers as a way to reduce undesirable acidity in cacao. However, longer storage periods are also associated with continued development or introduction of disease and contaminants that are highly undesirable in specialty cacao, and likely to be noticeable in the final product.Two post harvest treatments on measurements of brix, pH, pod weight, and seed with pulp weight in the pre-fermentation, early post-harvest processing stage were investigated, including cacao pods under storage treatments as well as separated by maturity under storage treatments. Fresh and over ripe fruits displayed the highest brix (17.53 average and 15.97 average, respectively) and overripe fruits with longer storage periods displayed the lowest (12.6 and 13.61, average respectively). Significant losses in brix, pod weight, and seed with pulp weight were

observed, and pH also significantly changed with storage treatments and ripeness. Post harvest treatments of pod storage and pod maturity significantly impact the pulp quality in early stages cacao processing, potentially influencing final cacao quality. When treatments are applied in order to reduce pulp acidity, shorter storage times appear to be equally effective in reducing pulp acidity, while also shortening the window for contamination and off flavor development. The relationship between pod maturity and pulp acidity begs further investigation for similar objectives.

3:15 PM - 3:30 PM (Wed. Sep 4, 2019 2:45 PM - 3:30 PM Room C)

[4-1445-C-03] Current Status of Monitoring Post-Harvest Potato Storage Units in Atlantic Canada

*Ahmad Al-Mallahi[1]　(1. Dalhousie University(Canada)）
Keywords: Storage Unit, Montioring System, Environmental Sensors, Potato tuber, Disease control

Potato is the largest crop in Atlantic Canada producing nearly one-third of the national share. Potato production in this region is characterized by short growing season and long storage periods where most of the produce destined for processing. Farmers, in this region, tend to have their own storage units where they pile in their yield immediately after the short harvesting season, while they gradually dispatch batches of their yield upon requests from processing factories. Storage lasts from autumn, through the harsh Canadian winter, until early summer during which farmers have to manage the storage units to keep their potatoes as fresh as possible and to avoid any spread of damage such as rots or other disease. The most common parameters to be monitored in a storage unit are temperature, relative humidity, and carbon dioxide concentration. These parameters control ventilation mechanism that pass air in underground ducts and through the potato pile. A mechanism of humidifying the air is occasionally actuated to increase humidity in the ventilating air to avoid tubers dehydration. Generally, the temperature is kept as low as 9 ℃ while the humidity is raised to as high as 95% to maintain optimum conditions. While sensors within the pile are usually used to monitor temperature, humidity sensors cannot sustain the storage conditions for one whole season. Therefore, farmers rely on portable sensors to measure humidity manually in a low frequency. Similarly, portable sensors are occasionally brought in to measure carbon dioxide concentrations which can be an indicator of disease breakout. Based on the current storage monitoring situation, temperature is fairly controlled by automatic temperature sensing and ventilators actuation, whereas humidity is controlled based on regular actuation of the humidification system and occasional review of the humidity levels. This situation can still cause misjudgment not only because of the low frequency of monitoring humidity but also the bias of measuring certain spots of temperature where the sensors are located. Since losing potatoes in the storage unit can be economically harmful, it is important to improve the current monitoring techniques to maintain potatoes fresh and minimize probability of disease breakout. In our research group at Dalhousie University, we are setting up a comprehensive plan to tackle problems associated with monitoring potato storage units. Our plan will include digging deep to find root causes of humidity sensor failure under storage conditions to develop more robust sensors. Also, we will rely on state-of-the-art technology of wireless sensing to have better understanding of the distribution of the parameters within the storage unit. These developments aim not only to improve the storage conditions but also to serve as early detection systems of disease breakouts. Considering the vast area that needs to be covered to collect data from different storage units scattered at different locations, we plan to develop remote data collection system to enable fast collection of information and centralized big data base for the region. The goal of big data analysis will be to find trends among the disconnected storage units and determine locations at higher risk of yield loss during storage.

Oral Session | Postharvest Machinery

[4-1445-D] Postharvest Machinery

Chair:Yukiharu Ogawa(Chiba University, Japan)
Wed. Sep 4, 2019 2:45 PM - 3:15 PM Room D (4th room)

[4-1445-D-01] **A Numerical Procedure for Supporting Garlic Root Trimming Machines Using Deep Learning Algorithms**

*Thuyet Quoc Dang[1], Morinobu Matsuo[1,2], Takeshi Haji[1], Tetsuo Kawaide[1], Yuichi Kobayashi[1]　(1. Institute of Agricultural Machinery, National Agriculture and Food Research Organization(Japan), 2. Central Region Agricultural Research Center, National Agriculture and Food Research Organization(Japan))

 2:45 PM - 3:00 PM

[4-1445-D-02] **A Nondestructive Acoustic Vibration System for Apple Firmness Assessment**

*Chengqiao Ding[1], Di Cui[1]　(1. Zhejiang University(China))

 3:00 PM - 3:15 PM

2:45 PM - 3:00 PM (Wed. Sep 4, 2019 2:45 PM - 3:15 PM Room D)

[4-1445-D-01] A Numerical Procedure for Supporting Garlic Root Trimming Machines Using Deep Learning Algorithms

*Thuyet Quoc Dang[1], Morinobu Matsuo[1,2], Takeshi Haji[1], Tetsuo Kawaide[1], Yuichi Kobayashi[1] （1. Institute of Agricultural Machinery, National Agriculture and Food Research Organization(Japan), 2. Central Region Agricultural Research Center, National Agriculture and Food Research Organization(Japan)）
Keywords: Garlic, Root trimming, Deep learning, Convolutional neural networks, Computer vision

Smart agricultural machinery is indispensable for modern postharvest processes for reducing human labor force, safety and increasing productivity. This study introduces a method to detect and evaluate the root trimming condition of garlics based on garlic images or the live streaming video from a personal computer webcam using convolutional neural network algorithms. This was an artificial intelligence system utilizing transfer learning techniques in deep learning. We aimed to develop a real-time classification system of garlic during the root trimming process and to provide signals to autonomously control a garlic trimming machine. The classification considered as three classes namely, good, bad and scratch. The good class consisted of successfully trimmed garlics whereas the bad class consisted of incompletely trimmed garlics which required further trimming. The scratch class consisted of defective garlics that should be removed during garlic postharvest processes. The classification system was automatically operated when a garlic was placed under the webcam. The analysis results were sent to two replays via serial ports for further automation processes. It effectively classified images for root trimming. The classification was instant, and its accuracy was about 96%. The signal can be used to develop an unmanned trimming machine. This system has the potential for high-impact applications in agricultural imaging, especially in postharvest machinery.

Oral Session | Postharvest Machinery

[4-1445-D] Postharvest Machinery

Wed. Sep 4, 2019 2:45 PM - 3:15 PM Room D (4th room)

[4-1445-D-02] A Nondestructive Acoustic Vibration System for Apple Firmness Assessment

*Chengqiao Ding[1], Di Cui[1] (1. Zhejiang University(China))

Keywords: Fruit firmness, Excitation device, Test parameters, Vibration characteristics

Fruit firmness is closely related to the physical structures and mechanical properties, which is an important index at different stages of the food supply chain. In this paper, a loudspeaker-based excitation device was designed and compared with the traditional vibration shaker. The apples were placed on a string bag and driven by the swept sine wave signals ranging from 50 to 2000 Hz. And the response signal of apples was acquired by a laser doppler vibrometer (LDV) which was hung on the top of the excitation units. The test parameters of detection system were optimized in the single factor experiment, and the superior combination of test parameters were as follows: the aperture of sound source was 40 mm, the distance between fruit surface and loudspeaker was 95 mm, and the posture style was that the apple was placed with its stem upward. After the optimization of detection system, six vibration characteristics were extracted from the frequency response function (FRF) to establish the relationship with fruit firmness obtained by the puncture test. The correlation results showed the stiffness of apples was closely related to elasticity index (EI) and stiffness coefficient (SC), which was considered as a dependent variable in different regression models. Furthermore, the highest correlation coefficient r_p of the prediction set was observed in the BP neural network model by using EI, the peak value at f_2 and the peak area as the independent variables (r_p=0.914, RMSEP=0.561 N mm^{-1}).

A Nondestructive Acoustic Vibration System for Apple Firmness Assessment

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ABSTRACT

Fruit firmness is closely related to the physical structures and mechanical properties, which is an important index at different stages of the food supply chain. In this paper, a loudspeaker-based excitation device was designed and compared with the traditional vibration shaker. The apples were placed on a string bag and driven by the swept sine wave signals ranging from 50 to 2000 Hz. And the response signal of apples was acquired by a laser doppler vibrometer (LDV) which was hung on the top of the excitation units. The test parameters of detection system were optimized in the single factor experiment, and the superior combination of test parameters were as follows: the aperture of sound source was 40 mm, the distance between fruit surface and loudspeaker was 95 mm, and the posture style was that the apple was placed with its stem upward. After the optimization of detection system, six vibration characteristics were extracted from the frequency response function (FRF) to establish the relationship with fruit firmness obtained by the puncture test. The correlation results showed the stiffness of apples was closely related to elasticity index (EI) and stiffness coefficient (SC), which was considered as a dependent variable in different regression models. Furthermore, the highest correlation coefficient r_p of the prediction set was observed in the BP neural network model by using EI, the peak value at f_2 and the peak area as the independent variables (r_p=0.914, RMSEP=0.561 N mm^{-1}).

Keywords: Fruit firmness Excitation device Test parameters Vibration characteristics

1. INTRODUCTION

With the increasing requirement for high-quality fruits, fruit classification and detection are becoming more and more important, which are based on both external and internal quality. External quality indicators mainly contain color, shape, size and appearance quality, while internal quality indicators mainly include chemical compositions (sugar, acidity, vitamin content, inorganic salt, ester, ethylene, etc.), texture and defects. As for the apple, firmness is widely used for its texture or ripeness evaluation, which is closely related to the physical structures and mechanical properties (Li et al., 2011; Pozrl et al., 2010). Accurate detection of firmness is indispensable in fruit supply chain. At harvest stage, firmness is utilized to determine the optimal harvest time and ripeness for edibility. In the grading process, firmness is the basis for classification. In the transportation, firmness is regard as a standard to select proper methods of transportation and packaging. During the storage, firmness helps to confirm storage temperature, humidity and time. As for the sale stage, firmness is used to assess the shelf-life and freshness, which deeply affect consumer purchase behaviors (Zhang et al., 2015). Fruit firmness detection methods can be divided into two classes, including destructive and nondestructive methods. The widely used destructive methods are the Magness-Taylor puncture test, which was deemed to be an industry standard. In the puncture test, a penetrometer records the force-deformation (F/D) curve by penetrating fruit tissue at a certain speed and extracts firmness indexes of fruit flesh based on F-D curve (Camps et al., 2005). However, destructive

methods are time-consuming, labor-intensive and local measurement. Thus, many nondestructive techniques have been developed for firmness assessment, such as acoustic vibration (Taniwaki and Sakurai, 2010), spectroscopy (Xing et al., 2006), ultrasonic (Mizrach and Flitsanov, 1999), etc. Among them, acoustic vibration method was commonly used in practice use, since it provides direct measurement of the mechanical and physical properties (García-Ramos et al., 2005; Grotte et al., 2002). Based on existing researches, a series of vibration characteristics were extracted to evaluate the fruit firmness, such as f^2m, $f^2m^{2/3}$ and $f^2m^{2/3}\rho^{1/3}$ (Abbott et al., 1992; Duprat et al., 1997; Schotte et al., 1999). To obtain the acoustic vibration characteristics, many measurement methods and experimental apparatuses were developed (Taniwaki and Sakurai, 2010). In order to not influence the original vibration of the sample, noncontact excitation devices and detection sensors were introduced to nondestructive measurement, such as the loudspeaker and the laser Doppler vibrometer (LDV). Muramatsu et al. (1996) used a small speaker to emit sound wave with frequencies from 200 to 2000 Hz to excite the fruits by an oscillator, and the response signal was acquired by a microphone on the opposite side. Similarly, Kataoka et al. (2016) developed a portable device to detect tomato firmness, which consisted of a smart phone, a microphone and speaker. The smart phone provided the swept sine signal from 20 to 10000 Hz in 1 s to excite fruit by speaker and captured the response signal by a microphone. Besides, the LDV is another alternative non-contact sensor to obtain the vibration velocity of the samples based on the Doppler shift of the laser beam frequency for its merits of noninterference movement, high spatial resolution, high precision and large measuring range (Murayama et al., 2006). In the early time, Muramatsu et al. (1997) applied the LDV system to monitor the firmness of apples, kiwifruits and pears. The results showed that vibration spectrum received by a laser doppler vibrometer had higher precision than the accelerometer method, especially in the frequency band from 800 to 1600 Hz. Lately, Abbaszadeh et al. (2013) developed a LDV system to estimate the firmness of watermelon. In the detection, the watermelon with a reflective film was excited by a mechanical shaker, and the vibration response signal was recorded by the LDV. The results showed that the prediction of stepwise multiple linear regression model (SMLR) based on phase spectrum was better than partial least squares regression model, and the determination coefficient of validation set was 0.9986.

The objectives of this research were to: (i) develop a loudspeaker-based excitation device and compare detection results with the traditional vibration shaker; (ii) to investigate the optimal test parameters in the single-factor experiment, including apple posture style, the aperture of sound source, and the distance between fruit surface and loudspeaker; (iii) to establish the relationships between apple firmness and vibration parameters in different regression models.

2. MATERIALS AND METHODS

2.1 Samples

'Fuji' apples (*Malus domestica* cv. Fuji, produced in Shanxi province, China) were purchased from the local fruit orchard, which has round shape, firm and juicy flesh, rich nutrition ingredients and good storage ability. A total of 48 apples with uniform size and spherical shape were selected and stored in the laboratory at 20 ℃ and 60 %RH. Before the test, each sample was placed at room temperature for 12 h and randomly coded. Then the morphological properties of each sample were measured three times, and average values were calculated for

analyses, including the mass (m), height (h) and equator diameter (d). After that, the vibration response signals and the firmness of samples were acquired by the following tests.

2.2 Vibration Response Signal Measurement

The design of the vibration measurement systems was shown in Fig.1, which was similar to the detection system used by Zhang et al. (2014) and Cui et al. (2015) (Fig.2). The system was mainly consisted of a loudspeaker (CS622C, Dayton Enterprises, USA), a microphone (40AE, M+P Enterprises, Germany), a LDV equipment (LV-S01, Sunny Instruments Singapore Pte., Ltd., Singapore), the NI data acquisition unit (USB-4431, National Instrument, Austin, USA), a power amplifier and a PC. In the measurement, the apple with a reflective film was placed on the string bag to vibrate freely. And the loudspeaker produced the swept sine wave signal (frequency range from 100 to 200 Hz in 1 s) to stimulate the apple. The sound signal was recorded by the microphone as the input signal (X_{in}). In the meantime, the LDV was used to acquire the vibration response signal from the fruit surface, which was regard as output signal (X_{out}). These two signals were changed from the time domain to the frequency domain based on Fast Fourier Transform (FFT), and the ratio was the frequency response function (FRF) (Fig.3). Then some vibration characteristics were extracted from FRF, such as the peak value (A), the second resonant frequency (f_2), the peak width at half height (w) and peak area ($S=Aw$).
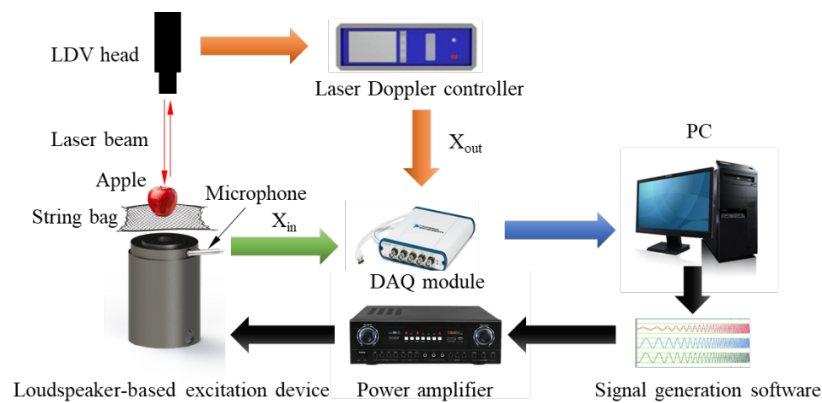
Figure 1. A loudspeaker-based LDV detection system.
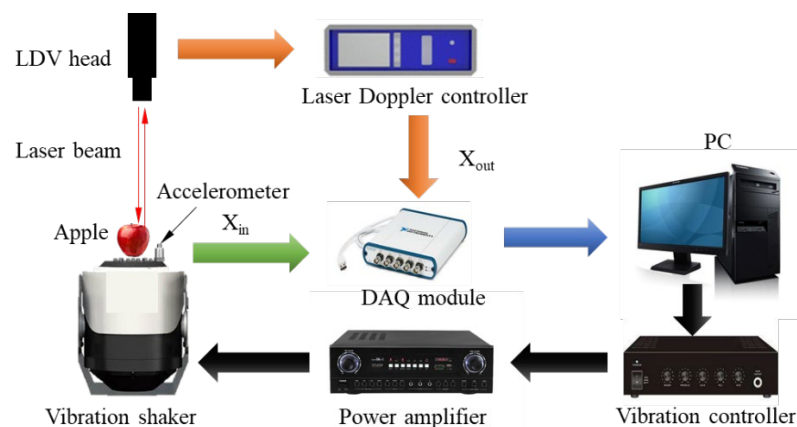
Figure 2. A vibration generator-based LDV detection system.
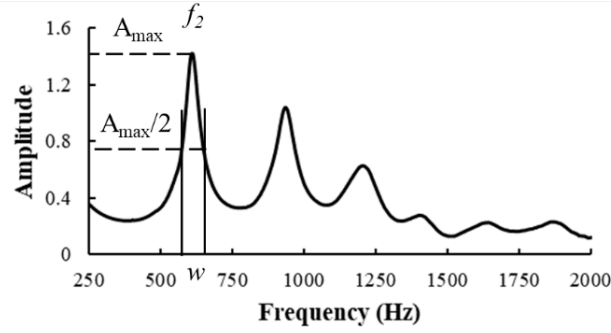
Figure 3. The typical FRF obtained from an apple by the acoustic vibration system.

2.3 Firmness Measurement

The firmness of apples was destructively measured by a standard penetrometer (TA-XT2i, Stable Micro Systems Ltd., England). In this study, three peeled detection points with equal intervals on the equator of the apple were selected. At each site, a flat-tip cylindrical probe (P/5) with a diameter of 5 mm was inserted into the sample. The penetration velocity and depth were 1 mm s^{-1} and 8 mm, respectively. Three firmness indexes were extracted from the force/deformation (F/D) curve, including stiffness (*Stif*, the slope of curve before the rupture point), *MT* firmness (the maximum force) and flesh firmness (*FF*, the mean force at a distance of 2–8 mm) (Fig.4). The average values of each firmness index were calculated and used for following analyses.
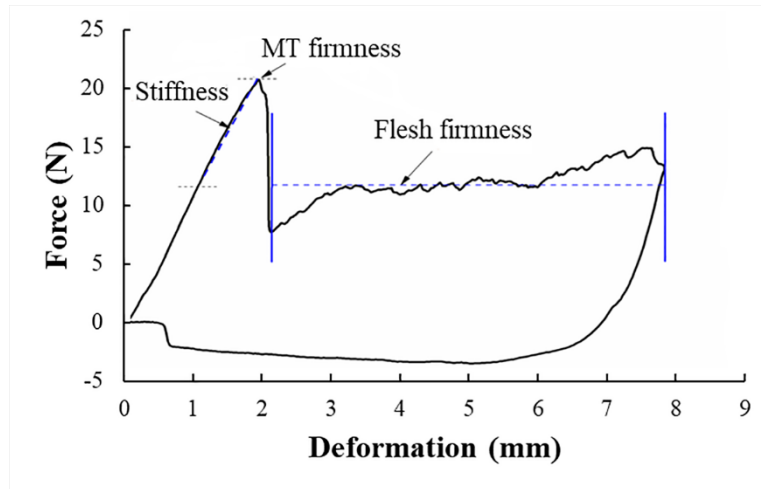
Figure 4. A representative force–deformation curve obtained from the puncture test.

2.4 Statistical Analysis

Correlation analysis was utilized to understand the direction and strength of the relationship between 2 individual variables (Cliff and Bejaei, 2018). In this study, the relationships among the firmness indexes extracted from force/deformation (F/D) curves and vibration characteristics obtained from FRF were assessed with values of the correlation coefficient (*r*), which were calculated through the Eq. (1).

$$r = \frac{\sum_{i=1}^{n}(x_i - \bar{x})(y_i - \bar{y})}{\sqrt{\sum_{i=1}^{n}(x_i - \bar{x})^2 \cdot \sum_{i=1}^{n}(y_i - \bar{y})^2}} \tag{1}$$

where x_i and y_i are nth measurements of variables X and Y (i=1,2, … , n), and $\bar{x}$ and $\bar{y}$ are the mean values of X and Y.

The repeatability of the vibration parameters was evaluated by coefficient of variation (CV), which was ratio of standard deviation (SD) of repeated measurements and their mean values (Mean). In the study, the value of CV below 10% showed that the detection system had a good repeatability (Wen et al., 2015).

Stepwise multiple linear regression (SMLR) method was a method to select significant independent variables and remove those that are not important based on the variance contribution in a linear regression model to avoid multicollinearity (Wang and Xie, 2014).

The partial least squares regression (PLSR) method was used to diminish the influence of high linear correlation between independent variables. In this study, the PLSR model was established with vibration characteristics from 48 apples. There were 32 and 16 samples in calibration and validation sets, respectively.

Support vector regression (SVR) was a supervised learning method which could be used for nonlinear regression analysis. The main characteristics of this method was to maintain the maximal margin and minimize the error (Liu et al., 2014).

Back propagation neural network (BPNN) was a multi-layer feedforward network trained by error inverse propagation algorithm, which was a supervised learning model. A neural network mainly consists of three parts: input layer, hidden layers, and output layer. Each layer had several neurons which was connected with other layers. The main characteristics were forward signal transmission and error back propagation. In the process of calculation, this method would adjust the network weights and threshold according to the prediction error until the result was close to the desired output (Liu et al., 2010).

3. RESULTS AND DISCUSSION

3.1 Physical Properties of Apples

Forty-eight fresh apples were selected with uniform size and shape in local orchard. The mean values and SD of the physical parameters such as mass, height, and equator diameter of test samples were presented in Table 1. It was revealed that the variations of these parameters were less, which indicated that the differences in their appearance were limited.

Table 1. Physical properties of experimental apples (n=48). (SD: Standard deviation)

Physical parameters	Mass (g)	Height (mm)	Equator diameter (mm)
Mean ± SD	280.06±31.41	79.47±4.28	74.53±3.78

3.2 Comparison of Contact and Noncontact Type Excitation Methods

The loudspeaker was used as a noncontact type device to excite the apple which was placed on a string bag, while the shaker was regard as a contact type device in excitation. The differences in performances of these two devices were compared based on the second resonant frequency (f_2) of 12 apples. Each measurement was repeated three times, and the deviation ratios (D) were utilized to describe the degree of the difference (Fig.5), which were calculated by the Eq. (2). The results showed that the f_2 obtained by shaker-based method was little higher than the

loudspeaker-based method. Due to the low values of deviation ratio, it was indicated that there was no significant difference in the second resonant frequency detection by these two excitation methods. However, the intensity of sound wave was relatively low, which may cause the insufficient excitation for large fruit.

$$D = \frac{f_2 - f_2'}{f_2} \qquad (2)$$

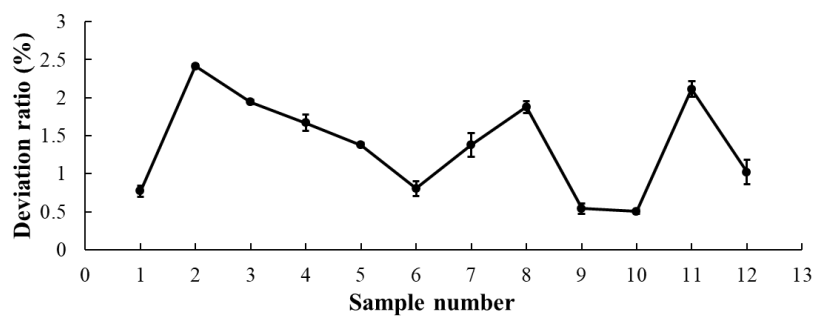where f_2 and f_2' were the second frequencies obtained by shaker-based method and loudspeaker-based method, respectively.

Figure 5. Deviation of the second resonant frequencies of the apple. The bars represent the standard error.

3.3 Repeatability of Vibration Parameters

The repeatability of vibration parameters was represented by the coefficient of variation (CV) of 12 apples (Fig.6). The results showed that the second frequency had the lowest CV value than other indexes. Besides, the peak value had better repeatability than the peak width at half height and the peak area. The CV values of all indexes were less than 10 %, which indicated that the loudspeaker was suitable for excitation in the detection.
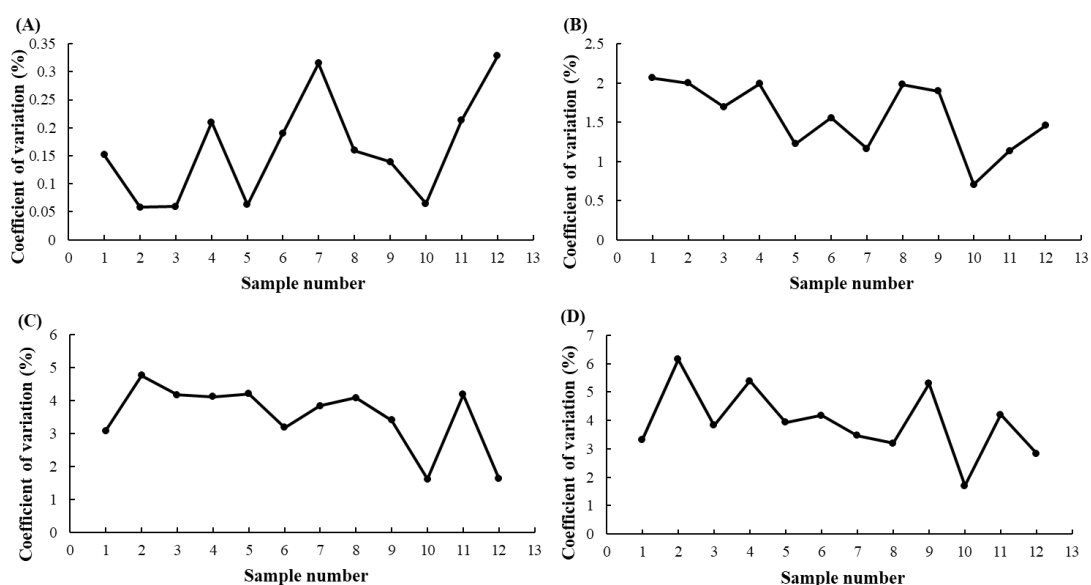
Figure 6. The coefficient of variation of the second resonant frequency (A), the peak value

(B), the peak width at half height (C) and the peak area (D).

3.4 Effects of Test Parameters on Vibration Signal
3.4.1 Different structural parameters
The schematic diagram of the loudspeaker-based excitation device was shown in Fig.7. In general, the intensity of the sound wave was closely related to the diameter of gasket and the distance between fruit surface and loudspeaker. In this section, the signal-to-noise ratio (SNR) was used to evaluate the performance of different structural parameters by single factor experiment.

Figure 7. The schematic diagram of the loudspeaker-based excitation device. (d: the diameter of gasket; h: the distance between fruit surface and loudspeaker)

The three sizes of gaskets were designed in this study, including 20 mm, 30 mm and 40 mm (Fig.8). The SNR values at different diameters of gaskets were shown in Fig.9. It was revealed that the 40 mm gasket obtained the largest SNR value, and there was no significant difference in the other two groups.
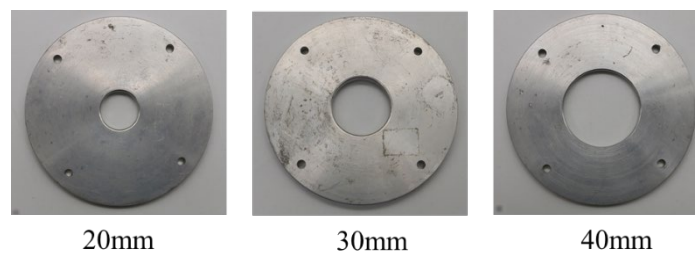
20mm   30mm   40mm
Figure 8. The different sizes of gaskets.
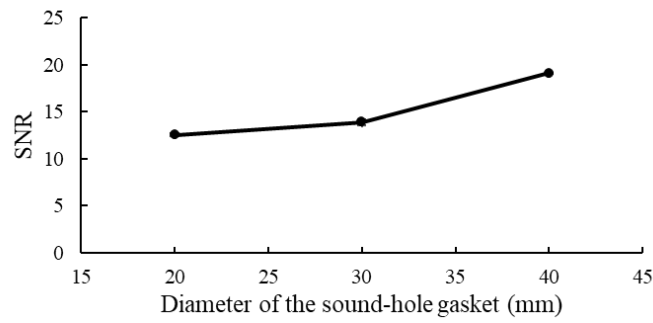
Figure 9. The SNR values of different sizes of gaskets.

The distance between fruit surface and loudspeaker could be adjusted from 95 mm to 155 mm. The SNR values at different distances were shown in Fig.10. The results showed that there was a nearly linear relationship between the SNR values and the distances. And the 95 mm group was found to have the maximum SNR value than other groups. Thus, the optimum size of the gasket and the distance were 40 mm and 95 mm, respectively.
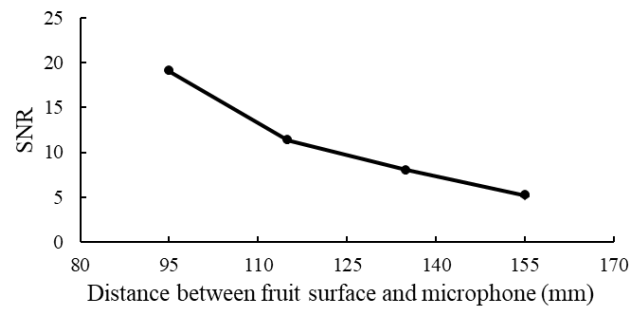
Figure 10. The SNR values of different distances between fruit surface and loudspeaker.

3.4.2 Different Detection Points and Posture Styles

The apple could be placed on the string bag in three different posture styles (Fig.11). In order to evaluate the repeatability of vibration parameters at each posture style, three detection points with equal intervals were selected as a group to compare. The performances of repeatability were represented by CV values (Fig.12). Good repeatability was found in each group of

detection points (CV<5%), which indicated that there was no significant difference in different

detection points at each posture styles. Besides, posture style B had high CV values in both resonant frequency and peak value.
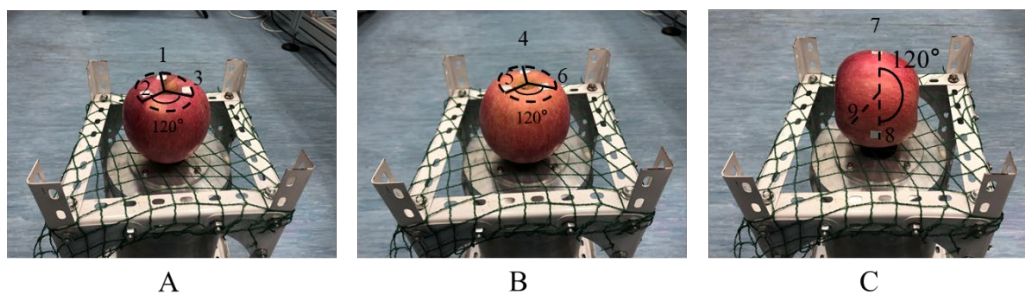
Figure 11. Different detection points and posture styles in detection system. A: the apple stem is upward; B: the apple calyx is upward; C: the apple stem-calyx is horizontal.
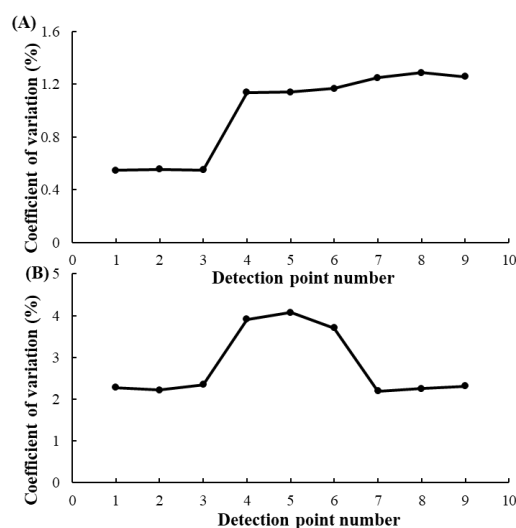
Figure 12. The coefficient of variation of resonant frequency (A), the peak value (B) in different detection points.

Fig.13 showed the SNR values at different posture styles. In general, the posture style A obtained the little higher SNR value than the posture style B. Besides, posture style C had lowest SNR value, which may be caused by unstable placements. Due to the biggest standard error at the posture style B, the posture style A was considered better and selected for the subsequent experiments.
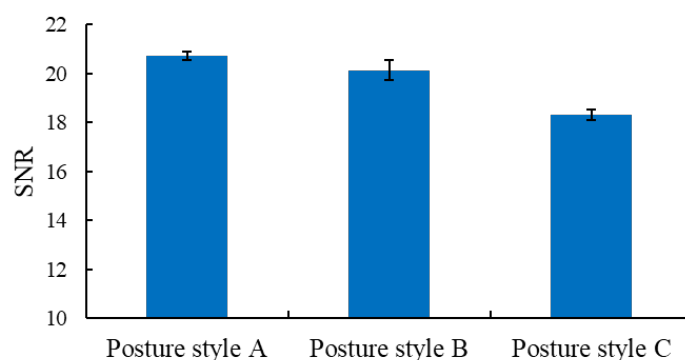
Figure 13. The SNR values of three posture styles. The bars represent the standard error.

3.5 Quantitative Analysis of Apple Firmness

After verification of the optimum test parameters, the vibration response signals of 48 apples were acquired. Four vibration characteristics were extracted from FRF, such as the peak value (A), the second resonant frequency (f_2), the peak width at half height (w) and peak area ($S=Aw$). Then elasticity index ($EI=f_2^2 m^{2/3}$) and stiffness coefficient ($SC=f_2^2 m$) were calculated to investigate the relationship with fruit firmness. In order to diminish the collinearity effect of these indexes on regression models, the inter-correlations of six variables were represented in Table 2. The results of correlation analysis demonstrated that the cross-correlations among the second resonant frequency, elasticity index (EI) and stiffness coefficient (SC) were closely correlated ($p \leq 0.01$), with r values between 0.484 and 0.827 (n = 48). Besides, w was strongly correlated with S ($r = 0.771$), EI ($r = -0.611$) and SC ($r = -0.641$). S was moderately correlated with A ($r = 0.544$), EI ($r = -0.459$) and SC ($r = -0.446$). The results also indicated that A was slightly correlated with the other four variables ($r = -0.075$ to -0.250), except S. Strong relationships among the independent variables would lead to the multicollinearity problem in the regression models. Thus, it was necessary to choose appropriate variables to improve the prediction of models. Due to the previous researches, the resonance frequency would be influenced by the object size. Thus, EI and SC were used to compensate for the difference in fruit size (Abbott et al., 1968; Cooke, 1972). In addition, f_2 and w would not be introduced as independent variables in the multiple regression model.

Table 2. Correlation coefficients (r) among vibration characteristics (n = 48).

Variable	f_2	A	w	S	EI	SC
f_2	1					
A	-0.307	1				
w	-0.075	-0.094	1			
S	-0.221	0.544*	0.771*	1		
EI	0.484**	-0.250	-0.611**	-0.499*	1	
SC	0.529**	-0.087	-0.641**	-0.446*	0.827**	1

Asterisks indicate statistical significance: ** significant correlation at the level of 0.01; *significant correlation at the level of 0.05.

The correlations between the firmness indexes obtained by the puncture test and vibration characteristics (EI and SC) were showed in Table 3. The results revealed that stiffness had the highest correlation with EI and SC, which was regard as a dependent variable in regression models.

Table 3. Correlation coefficients (r) among vibration characteristics and firmness indexes.

Variable	Stif	MT	FF
EI	0.852**	0.534*	0.222
SC	0.629**	0.434*	0.242

Asterisks indicate statistical significance: ** significant correlation at the level of 0.01; *significant correlation at the level of 0.05.

The results of the unary linear regression models were showed in Table 4. All factors (EI, SC, f_2, w, S), except A, were strongly correlated with fruit stiffness. The best unary regression model for prediction of stiffness was established by using EI as an independent variable, whose r_p and RMSEP of prediction set were 0.830 and 0.770, respectively.

Table 4. Statistical results of the unary linear regression models for determining stiffness of apples.

Factors	Regression model	Calibration set		Prediction set	
		r_c	RMSEC	r_p	RMSEP
EI	$y = 4.643 \times 10^{-5}x + 2.768$, F=48.824 (**$P<0.01$)	0.894	0.556	0.830	0.770
SC	$y = 3.961 \times 10^{-5}x + 7.740$, F=13.090 (**$P<0.01$)	0.694	0.628	0.611	1.094
f_2	$y = 0.010x + 5.837$, F=8.016 (**$P<0.01$)	0.556	0.487	0.517	1.182
A	$y = -69.934x + 15.082$, F=1.011 ($P>0.05$)	0.226	1.198	0.210	1.351
w	$y = -0.031x + 16.769$, F=11.078 (**$P<0.01$)	0.573	0.738	0.579	1.126
S	$y = -1.373x + 16.295$, F=12.149 (**$P<0.01$)	0.745	0.540	0.597	1.109

Asterisks indicate statistical significance: ** significant correlation at the level of 0.01; *significant correlation at the level of 0.05.

In order to diminish the collinearity effect of vibration characteristics on a multiple linear regression model, stepwise multiple linear regression (SMLR) was utilized to variable selection. The performances of SMLR models were showed in Table 5. It could be seen that using EI, the peak value and the peak area could obtain the better prediction result than another model, and the r_p and RMSEP of prediction set were 0.871 and 0.712 N mm^{-1}, respectively.

Table 5. Statistical results of SMLR model for determining stiffness of the apples.

(I)

Factors	Regression coefficient	Calibration set		Prediction set	
		r_c	RMSEC	r_p	RMSEP
EI	4.042×10^{-5}				
A	22.145				
S	-0.0472	0.909	0.474	0.871	0.712
Constant	4.643				
	F=16.210, **$P<0.01$				

(II)

Factors	Regression coefficient	Calibration set		Prediction set	
		r_c	RMSEC	r_p	RMSEP
SC	2.601×10^{-5}				
A	9.716				
S	-0.934	0.795	0.600	0.701	1.044
Constant	11.352				
	F=6.173, **$P<0.01$				

Asterisks indicate statistical significance: ** significant correlation at the level of 0.01; *significant correlation at the level of 0.05.

The performances of different nonlinear models for prediction of stiffness were represented in Table.6. Compared with the results of the unary linear regression model, PLSR and BP neural network model had the better prediction ability. Besides, SVR model was the worst in calibration set and prediction set. Furthermore, the highest correlation coefficient r_p of the prediction set was obtained in the BP neural network method by using EI, A and S as independent variables ($r_p = 0.914$, RMSEP = 0.561 N mm^{-1}).

Table 6. Results of quantitative analysis of stiffness by different nonlinear models.

Modeling method	Input variables	Calibration set		Prediction set	
		r_c	RMSEC	r_p	RMSEP
PLSR	EI, A and S	0.904	0.557	0.842	0.754
	SC, A and S	0.727	0.949	0.688	1.089
SVR	EI, A and S	0.893	0.519	0.801	0.671
	SC, A and S	0.699	0.627	0.568	0.994
BP neural network	EI, A and S	0.957	0.413	0.914	0.561
	SC, A and S	0.889	0.617	0.858	0.805

4. CONCLUSION

The loudspeaker-based excitation device was designed and used in the LDV detection system. The test parameters of detection system were optimized based on the results of CV values and SNR values under different test conditions. A better combination of test parameters for vibration response signal measurement were as follows: the aperture of sound source was 40 mm, the distance between fruit surface and loudspeaker was 95 mm, and posture style was that the apple was placed with its stem upward. Based on optimized system, the vibration responses of 'Fuji' apples were acquired, and then six vibration characteristics were extracted, including the peak value, the second resonant frequency, the peak width at half height, peak area, EI and SC. The correlations between the firmness indexes obtained by the puncture test and vibration characteristics (EI and SC) were revealed that stiffness had better performance than other firmness indexes, which was regard as a dependent variable in different regression models. Moreover, the best prediction of firmness was observed in the BP neural network model by using EI, A and S as input variables, and the correlation coefficient r_p of the prediction set was 0.914 and RMSEP was 0.561 N mm^{-1}.

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Oral Session | Postharvest/Food Technology and Process Engineering

[4-1600-A] Postharvest/Food Technology and Process Engineering (3)

Chair:Lotis Escobin Mopera(University of the Philippines Los Banos, Philippines), Natthawuddhi Donlao(Mae Fah Luang University, Thailand)

Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A (Main Hall)

[4-1600-A-01] **Electricity Production from Xylose in Microbial Fuel Cells Started with Three Different Inoculum Sources**

*Yite Liu[1], Megumi Ueda[1], Tadashi Chosa[1], Seishu Tojo[1] （1. Tokyo University of Agriculture and Technology (Japan)）

4:00 PM - 4:15 PM

[4-1600-A-02] **Biodegradable Food Packaging from Cavendish Banana (*Musa acuminata*) Peduncle Fiber**

Kittaporn Ngiwngam[1], Nor Jihan Jantan[2], *Wirongrong Tongdeesoontorn[1,3] （1. School of Agro-Industry, Mae Fah Luang University, Chiang Rai 57100 (Thailand), 2. School of Industrial Technology, Universiti Teknologi MARA, Shah Alam, Selangor 42300 (Malaysia), 3. Research Group of Innovative Food Packaging and Biomaterials, Mae Fah Luang University, Chiang Rai, 57100 (Thailand)）

4:15 PM - 4:30 PM

[4-1600-A-03] **Assessment of the Physical Characteristics of Maize (Zea mays) stored in different Positions within the Metallic Silos**

*BABATOPE ALBERT ALABADAN[1], CALLISTUS A. OKOLO[2] （1. Federal University, Oye Ekiti (Ikole Ekiti Campus)(Nigeria), 2. Federal University of Technology, Minna(Nigeria)）

4:30 PM - 4:45 PM

[4-1600-A-04] **Rice Analogue: Technology for Rice Enrichment and Food Diversification**

*Lerjun Monilla Penaflor[1] （1. Food Engineering Division, Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Banos (Philippines)）

4:45 PM - 5:00 PM

[4-1600-A-05] **Optimization of Process Conditions for *Batuan* [*Garcinia binucao* (Blanco) Choisy] Fruit Powder Production**

*Al Kaxier Guzman Ancheta[1], Erlinda I. Dizon[2] （1. University of the Philippines Los Banos(Philippines), 2. University of the Philippines Los Banos (Philippines)）

5:00 PM - 5:15 PM

[4-1600-A-06] **Effect of Processing Conditions on Bioactive Compounds and Antioxidant Activities of Tea Infusion**

*Wei Qin[1], Sunantha Ketnawa[1], Florencio, Jr. Collado Reginio[1,2], Ryutaro Yamada[3], Takuya Araki[3], Yukiharu Ogawa[1] （1. Graduate School of Horticulture, Chiba University.(Japan), 2. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Baños(Philippines), 3. Institute of Fruit Tree and Tea Science, NARO(Japan)）

5:15 PM - 5:30 PM

[4-1600-A-07] *In Vitro* **Release Characteristics of Sugars and Hydrolysis of Starch During Simulated Digestion of Saba banana at Different Maturity**

Stages

*Florencio, Jr. Collado Reginio[1,2], Wei Qin[1], Yukiharu Ogawa[1] （1. Graduate School of Horticulture, Chiba University(Japan), 2. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Bañ os(Philippines)）

5:30 PM - 5:45 PM

[4-1600-A-08] *In Vitro* Examination of Starch Digestibility and Antioxidant Activities of Amaranth Grains

*Xiaoyan Xiong[1], Florencio Jr. Collado Reginio[1,2], Sukanya Thuengtung[1], Sunantha Ketnawa[1], Yukiharu Ogawa[1] （1. Graduate School of Horticulture, Chiba University(Japan), 2. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Baños(Philippines)）

5:45 PM - 6:00 PM

[4-1600-A-09] Effects of Cell Structure Changes of Citrus Peel on the Digestibility of Intracellular Antioxidants during *in vitro* Digestion

*Yidi Cai[1], Yukiharu Ogawa[1] （1. Graduate School of Horticulture, Chiba University(Japan)）

6:00 PM - 6:15 PM

Oral Session | Postharvest/Food Technology and Process Engineering
[4-1600-A] Postharvest/Food Technology and Process Engineering (3)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A (Main Hall)

[4-1600-A-01] Electricity Production from Xylose in Microbial Fuel Cells Started with Three Different Inoculum Sources

*Yite Liu[1], Megumi Ueda[1], Tadashi Chosa[1], Seishu Tojo[1]　(1. Tokyo University of Agriculture and Technology (Japan))

Lignocellulosic biomass from agricultural residues is considered as a promising feedstock for the productions of bioethanol. However, the conversion of bioethanol fermentation from lignocellulosic biomass is limited since its low efficiency of utilization of xylose. Microbial Fuel Cells (MFCs), the bioelectrochemical systems that use microorganisms as biocatalysts to oxidize organic and inorganic matters and recover electrons, shows the possibility to degrade xylose and generate electricity directly. This study aimed to investigate how the substrate was degraded and characteristics of electricity generation were influenced by comparing 3 different inoculum sources in the MFCs using xylose and acetate (reference) as sole carbon source. Six membrane-less MFCs with a single chamber and air-cathode in total volume of 26mL were inoculated with 3 inoculum sources, methane fermentation broth, cow dung compost and anaerobic sludge from a sewage treatment facility, respectively with two repetitions. The voltage across an external electric resistor in the circuit of the MFC was measured with a data logger at 5 minutes interval. Concentrations of xylose and volatile fatty acids were analyzed by HPLC after the depletion of carbon source. Polarization curves were made by varying the external resistance from 10 ohms to 1000 ohms. Coulombic Efficiency (CE) was calculated according to reduced carbon source and produced total coulombs by integrating the current over time. Results showed that all the three inoculums contained electrogenic bacteria, and the MFCs produced steady electricity from both acetate and xylose, whereas the characteristics of voltage output, substrate degradation and CE are different.

Electricity Production from Xylose in Microbial Fuel Cells Started with Three Different Inoculum Sources

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ABSTRACT

Lignocellulosic biomass from agricultural residues is considered as a promising feedstock for the productions of bioethanol. However, the conversion of bioethanol fermentation from lignocellulosic biomass is limited since its low efficiency of utilization of xylose. Microbial Fuel Cells (MFCs), the bioelectrochemical systems that use microorganisms as biocatalysts to oxidize organic and inorganic matters and recover electrons, shows the possibility to degrade xylose and generate electricity directly. This study aimed to investigate how the substrate was degraded and characteristics of electricity generation were influenced by comparing 3 different inoculum sources in the MFCs using xylose and acetate (reference) as sole carbon source. Six membrane-less MFCs with a single chamber and air-cathode in total volume of 26mL were inoculated with 3 inoculum sources, methane fermentation broth, cow dung compost and anaerobic sludge from a sewage treatment facility, respectively with two repetitions. The voltage across an external electric resistor in the circuit of the MFC was measured with a data logger at 5 minutes interval. Concentrations of xylose and volatile fatty acids were analyzed by HPLC after the depletion of carbon source. Polarization curves were made by varying the external resistance from 10 ohms to 1000 ohms. Coulombic Efficiency (CE) was calculated according to reduced carbon source and produced total coulombs by integrating the current over time. Results showed that all the three inoculums contained electrogenic bacteria, and the MFCs produced steady electricity from both acetate and xylose, whereas the characteristics of voltage output, substrate degradation and CE are different.

Keywords: Microbial fuel cell Xylose Power generation Biomass energy

1. INTRODUCTION

With the depletion of global fossil fuels and environmental pollution, renewable energy has received widespread attention increasingly (Mäkinen et al., 2013). Since its abundance, low pollution and great benefit for human society, ethanol production from lignocellulosic biomass became one of the most promising alternatives to replace traditional fossil energy.

However, the economical bio-ethanol production would be achieved only if both hexose and pentose sugars from lignocellulosic biomass are converted to ethanol efficiently. Although significant improvements such as recombination have been made in the microorganisms for improving the efficiency of converting pentose sugars into ethanol, the bioconversion of pentoses to ethanol is still one of the major bottlenecks for practical application (Kuhad et al., 2011).

Xylose, the most representative pentose, also the second most abundant carbohydrate after glucose from lignocellulose hydrolysate (Rubin, 2011), has been investigated in few studies. Those studies indicated stirring, initial concentration, fed-batch or continuous-flow, temperature, and use of electron mediator, have influence on the power generation of the xylose-fed MFCs (Huang et al., 2008a; Huang et al., 2008b; Dessì et al., 2018; Thygesen et al., 2009). However, the influences from different inoculums have not been published before.

In this study, to find better inoculums for electricity generation from xylose, characteristics of substrate degradation and electricity generation were investigated in the xylose-fed MFCs starting with three different inoculums.

2. MATERIALS AND METHODS

2.1 Inoculums

Three different inoculums describe below were inoculated to the duplicate MFCs respectively. (i) Methane fermentation broch from the biomass plant, Ogawa-machi, Saitama, Japan. (ii) Cow dung

compost from FS Center, TUAT, Fuchu-shi, Tokyo, diluted with 3 times volume of distilled water, and then filtered with a net before inoculating. (iii) Anaerobic sludge from North Tama Water Treatment Center, Fuchu-shi, Japan, without pretreatment. All the inoculums were stored in a refrigerator at 4°C.

2.2 MFC construction and operation

Six membrane-less MFCs with a single chamber and air-cathode in total volume of 26 mL were used in this study. Carbon cloth (TORAYCA cloth CO6343, TORAY, Japan) was used as anode after treated in a muffle furnace at 450 °C for 30min. The air-cathode was made of carbon cloth (EC-CC1-060T, ElectroChem, Inc, America) with a platinum (0.5 mg/cm^2) catalyst layer on the water side, and four layers of polytetrafluoroethylene (PTFE) diffusion were coated on the air side to prevent water (Cheng et al., 2006). Anode and cathode (reaction surface area of 9.62 cm$^{2)}$ were placed on the front and rear sides of the chamber, connecting with two twisted titanium wires served as current collectors. In order to change medium, 2 holes were drilled on the left and right side of the MFC respectively.

The medium containing was as follows (per liter): 0.31 g NH$_4$Cl, 0.13 g KCl, 2.54 g NaH$_2$PO4, 10.32 g NaH$_2$PO$_4$·12H$_2$0, 5 ml MEM Vitamin, 12.5 ml mineral solution. The mineral solution containing was as follows (per litter): 3 g MgSO$_4$·7H$_2$O, 0.5 g MnSO$_4$·H$_2$O, 1 g NaCl, 0.1 g FeSO$_4$·7H$_2$O, 0.1 g CoCl$_2$·6H2O, 0.1 g CaCl$_2$, 0.1 g ZnSO$_4$·7H$_2$O, 0.01 g CuSO4·5H$_2$O, 0.01 g H$_3$BO$_3$, 0.01 g Na$_2$MoO$_4$·2H$_2$O and 0.02 g NiCl$_2$·6H$_2$O. Sodium acetate (1g/L, control) or xylose (1g/L) as carbon source.

The inoculum sources were mixed with sodium acetate medium (v/v=1:1) for the initiation of the MFCs and replaced 2 or 3 times to form the bio-film on anode surface. After obtaining stable electricity generation, the mixed medium was changed to sodium acetate medium then xylose medium. Medium was fully discarded and refilled each time when the voltage decreased to under 0.05 V. Before polarization curve and measurement of organic acid or xylose, mediums were changed at least 3 times. All MFCs were operated at 30 °C in an incubator under fed batch conditions.

2.3 Analytical methods

MFCs connected with an external electric resistance (1000 ohms) in parallel, and the voltage across the resistance was recorded every 5 minutes by a data logger (GL240, GRAPHTEC, Japan).

Coulombic efficiency (CE), which defined as the fractional recovery of electrons from the substrate, was calculated by the following equation (Oh et al., 2004).

$$CE = \frac{M_s \int_0^t I dt}{F b_{es} V_{An} \Delta c} \qquad (1)$$

where, M_s is the molecular weight of substrate, t is the run time from medium replacement to the depletion of substrate, I is the current calculated as $I = V/R$, F is Faraday's constant, b_{es} is the number of electrons exchanged per mole of substrate (Accounting 20 mol electrons exchanged per mol of xylose and 8 mol for acetate), V_{An} is the volume of the MFC, Δc is the replacing time in the substrate over time t.

For calculating the maximum power density, polarization curves were made by varying the external resistance (Open circuit, 1000, 510, 240, 100, 51, 10 ohms). Power was calculated as $P = V^2/R$ and power density was calculated by the area of the anode.

After at least 3 times of replacing medium, the medium of MFCs were sampled at the end of batch cycle (Voltage < 0.05V). Time of depletion of one cycle was calculated from medium replacing until the voltage decrease under 0.05V.

Organic acids decomposed from xylose (Huang et al., 2008a) were performed on lactic acid, formic acid, acetic acid, propionic acid by HPLC (Shimadzu, UFLC Prominence) with column: Shim-pack SCR-102H (Shimadzu GLC) and detector: CDD-6A (Shimadzu). Xylose was measured by HPLC (Shimadzu, UFLC Prominence) with column: Shim-pack ISA-07/S2504 (Shimadzu GLC) and detector: RF-10AXL. All the operations were measured according to the instrument manufacturer's protocol.

3. RESULTS AND DISCUSSION

3.1 Electricity generation characteristics

After several cycles (3 cycles fed by sodium acetate or 6 cycles fed by xylose), the duplicate MFCs showed similar trends of electricity generation (maximum voltage, maximum power density,

corresponding resistance of maximum power density, CE and time of depletion) for methane fermentation broth-inoculated MFCs (Methane1 and Methane2) and sludge-inoculated MFCs (Sludge1 and Sludge2), while cow dung compost-inoculated MFCs (Compost1 and Compost2) were slightly different (Table 1).

Table 1. Electricity generation characteristics of MFCs fed by sodium acetate and xylose.

Substrate	Characteritics	Methane1	Methane2	Compost1	Compost2	Sludge1	Sludge2
Sodium Acetate (1g/L)	Maximum voltage (V)	0.540	0.552	0.528	0.574	0.571	0.571
	Maximum power density (mW/m^2)	718.93	675.86	493.33	648.61	745.86	710.35
	Corresponding resistance of maximum power density (ohms)	100	100	510	240	240	240
	CE (%)	25.96	23.46	15.54	17.53	16.08	17.69
	Time of depletion (h)	41.17	37.50	25.00	30.17	30.83	29.25
Xylose (1g/L)	Maximum voltage (V)	0.556	0.573	0.554	0.575	0.581	0.592
	Maximum power density (mW/m^2)	665.48	662.09	442.56	605.77	720.91	774.64
	Corresponding resistance of maximum power density (ohms)	240	240	510	240	240	100
	CE (%)	16.44	17.43	15.12	16.96	16.64	18.69
	Time of depletion (h)	40.67	39.83	34.17	40.42	42.75	43.92

3.2 Voltage

The voltage of the last cycle fed by sodium acetate and the sixth cycle fed by xylose were showed in Figure 1, and the initial six cycles fed by xylose (except for the cycle for measuring power density) was showed in Figure 2.
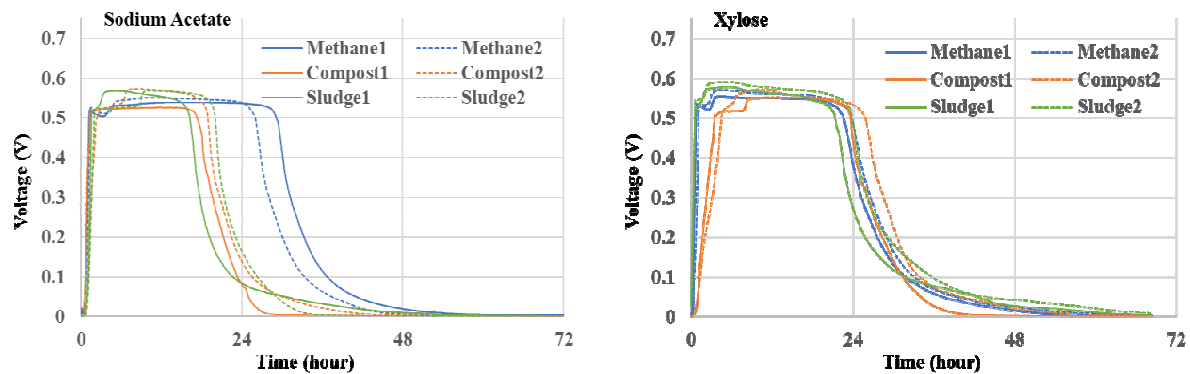
Figure 1. Voltage by time. Last cycle fed by sodium acetate and the 6th cycle fed by xylose.
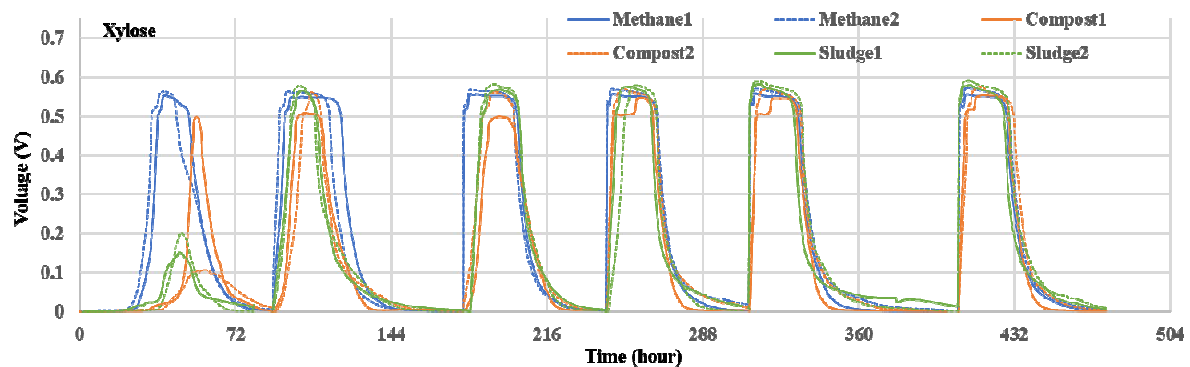
Figure 2. Voltage of the initial six cycles fed by xylose.

3.3 Adoption of xylose

As the increment number of cycles, voltage, time of depletion and CE became similar in all the MFCs fed by xylose (Figure 2 & Figure 3). Time of depletion was decreased, and CE was increased in the 6 cycles. Time of depletion ranged from 55.3 to 74.2 hours in the first cycle while it ranged from 34.9 to 43.8 hours in the sixth cycle. CE ranged from 3.03 % to 14.49 % in the first cycle while it ranged from 15.12 % to 18.69 % in the sixth cycle. Methane fermentation broth-inoculated MFCs showed shortest adaptation time, reached comparatively high CE and voltage almost at the first cycle with short lag time.
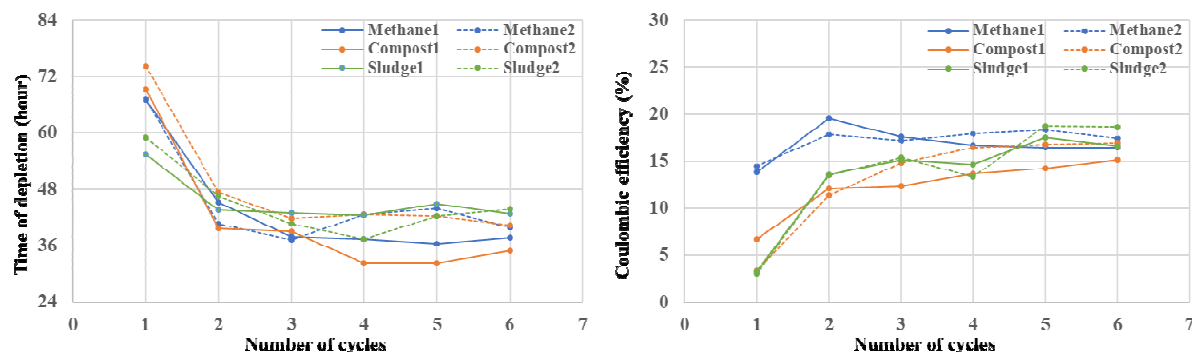
Figure 3. Time of depletion and coulombic efficiency changing by number of cycles.

3.4 Performance assessment of MFCs

According to the last cycle of sodium acetate and the sixth cycle of xylose, the performances of MFCs were as shown in Figure 4. Fed by xylose, sludge-inoculated MFCs had the highest of maximum voltage, maximum power density and CE although the longest time of depletion. Comparing with the methane fermentation broth-inoculated MFCs and sludge-inoculated MFCs respectively, the maximum voltage was 3.8 % and 3.8 % higher, the maximum power density was 11.2 % and 29.9 % higher, the CE was 4.1 % and 9.2 % higher, and the time of depletion was 7.1 % and 13.9 % longer. The performance of methane fermentation broth-inoculated MFCs were slightly lower than sludge-inoculated MFCs, however they had much higher CE when fed by sodium acetate. Cow dung compost-inoculated MFCs had the lowest maximum power density and CE, and it is possibly due to the different performance of the duplicate MFCs.
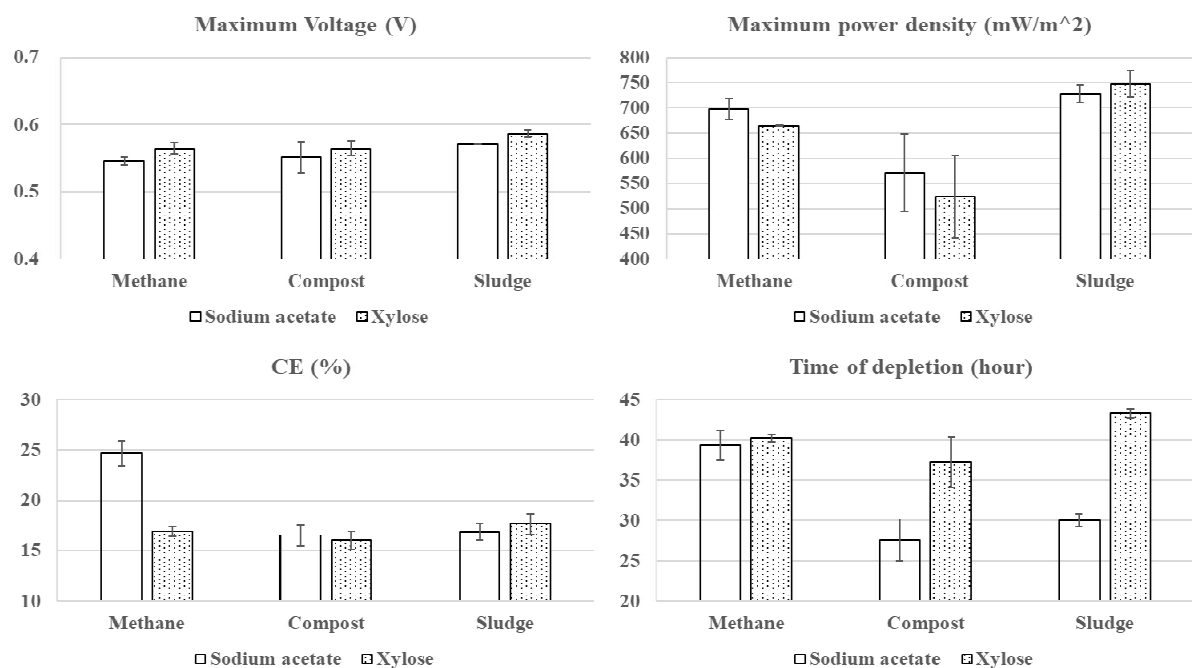
Figure 4. Performances of MFCs inoculated by different inoculum sources.

3.5 Substrate degradation
After at least 3 cycles of medium replacement, xylose and organic acid (lactic acid, formic acid, acetic acid, and propionic acid) were not detected by HPLC at the end of cycle (Voltage below 0.05V).

4. CONCLUSION
In this study, the characteristics of electricity generation was investigated in xylose-fed MFCs starting with three different types of inoculum sources, methane fermentation broth, cow dung compost and anaerobic sludge. Sludge-inoculated MFCs had the highest performance of electricity generation while methane fermentation broth-inoculated MFCs had the shortest adaptation time for xylose.

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4:15 PM - 4:30 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A)

[4-1600-A-02] Biodegradable Food Packaging from Cavendish Banana (*Musa acuminata*) Peduncle Fiber

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Keywords: Cavendish banana, Banana peduncle, Fiber, Biodegradable food packaging, Waste utilization

This study deals with the determination of chemical composition and the study of the pulping potentialities of banana peduncle waste in Chiang Rai, Thailand to produce biodegradable food packaging. Before the pulping process, chemical composition of banana fiber (BF) was identified. The result showed that holocellulose content in banana peduncle was 54.6±4.42 % (w/w) dried sample then lignin content was 14.42±0.39% dried sample. The banana fiber consisted of alpha-, beta-, and gamma-cellulose contents 59.96±0.03%, 2.89±1.05%, 37.35±1.3% (w/w) of holocellulose, respectively. After that, the molded pulp tray from banana peduncle fiber was formed by using the hydraulic hot-pressing machine at 140 ˚C, 250 psi for 7 minutes. Dried fiber around 15 g was used for the tray forming and the properties of the tray were determined. For color measurement, lightness (L*) of pulps were decreased from 46.47 to 38.18 by increasing NaOH concentration. Grammage of molded pulp trays from 10, 15, and 20% NaOH were 632.40± 25.37, 705.11±30.58, and 689.13±79.86 g/m², respectively. Tensile index and compression index of molded pulp tray at 10% NaOH showed the highest value as 22.12±1.23 MPa.m²/g and 25.03±1.04 MPa.m²/g. The tray at 20% NaOH gave the highest value of thickness swelling at 37.14±2.67%. The results indicated that the BF molded pulp tray from pulping process with 10% NaOH has good properties as high tensile index, compression index, MOR, and water wetting time which can be used as biodegradable food packaging.

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1600-A] Postharvest/Food Technology and Process Engineering (3)

Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A (Main Hall)

[4-1600-A-03] Assessment of the Physical Characteristics of Maize (Zea mays) stored in different Positions within the Metallic Silos

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Keywords: Maize, Storage, Metallic silos, Physical, Characteristics

The assessment of the moisture content (MC), hectoliter weight (HW), insect damage (ID), broken grains (BG), mould infestation (MI), viability/germinability (VG) and foreign matters (FM) of maize in different positions in the metallic silos during eight months storage was presented in this study. Seven and two positions in the maize bulk were investigated in the 250 MT and 1 MT silos respectively. The initial values obtained were compared with the values obtained during storage using statistical packages. The levels of significance for the variables are MC, $10.0\pm0.57\%$; HW, $72.9\pm0.10\%$; ID, $0.29\pm0.03\%$; BG, $0.55\pm0.01\%$; MI, $0.00\pm0.02\%$; VG, $100\pm0.00\%$ and FM, $0.80\pm0.60\%$ in respect to positions of the grains in the bulk. Variables were all significant (<0.05) except MC, HW and FM irrespective of the size of the metallic silos. The mean deviations of the variables from the control decreased for MC, HW, BG, VG and FM while ID and MI increased with storage period. The physical characteristics of maize were not influenced by the size of silo. The 1MT silo silo performed better in VG while bigger silo efficiency depends on the position of the grains in the bulk.

ASSESSMENT OF THE PHYSICAL CHARACTERISTICS OF MAIZE *(Zea mays)* STORED IN DIFFERENT POSITIONS WITHIN THE METALLIC SILOS

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ABSTRACT

The assessment of the moisture content (MC), hectolitre weight (HW), insect damage (ID), broken grains (BG), mould infestation (MI), viability/germinability (VG) and foreign matters (FM) of maize in different positions of the metallic silos during eight months storage was presented in this study. Seven and two positions in the maize bulk were investigated in the 2500MT and 1MT silos respectively. The initial values obtained were compared with the values obtained during storage using statistical packages. The levels of significance for the variables are MC $10.0\pm0.57\%$, HW $72.9\pm0.10\%$, ID $0.29\pm0.03\%$, BG $0.55\pm0.01\%$, MI $0.00\pm0.02\%$, VG $100\pm0.00\%$ and FM $0.80\pm0.60\%$ in respect to positions of the grains in the bulk. Variables were all significant ($p<0.05$) except MC, HW and FM irrespective of the size of the metallic silos. The mean deviations of the variables from the control decreased for MC, HW, BG, VG and FM while ID and MI increased with storage period. The physical characteristics of maize were not influenced by the size of silo. The 1 MT silo performed better in VG while bigger silo efficiency depends on the position of the grains in the bulk.

Keywords: Maize, storage, metallic silos, physical characteristics

4:45 PM - 5:00 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A)

[4-1600-A-04] Rice Analogue: Technology for Rice Enrichment and Food Diversification

*Lerjun Monilla Penaflor[1] (1. Food Engineering Division, Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Banos (Philippines))
Keywords: rice analogue, rice enrichment, food diversification, cereal grains, tubers crops

An optimization process for making rice-like kernels or rice analogue through cooking the fusion of different cereal grains and tuber crops mixed with water to produce a partially gelatinized mixture then extruded forming a rice shaped kernel and finally dried and cooled. The objective of the study is to develop a technology for reconstituting broken milled rice and explore the potentials of other cereal grains and tuber crops to enhance the nutritional composition and physico-chemical properties of the rice analogue specifically investigate its possibilities as a medium for rice enrichment and food diversification.The fusion of different cereal grains and tubers crops is suitable approach for enhancing the nutritional composition and physicochemical properties of the rice analogue. The products meets the nutritional criteria and could be developed as a medium for rice enrichment, as well as a food diversification for billion people. It deals an innovative technique of utilizing locally available grains and carbohydrate sources into a new staple food, but appropriate processing technology and comprehensive research is essential for developing rice analogue with specific functional properties, such as low glycemic index, high protein and fiber content. An appropriate medium should be widely adopted, but not require changes to daily healthy metabolism at the same time makeable from a wide range of sources. Rice analogue has the potential to provide for an observable gap in current fortification systems. The establishment of a new model offers a novel way to encourage people to food diversification and promote rice analogue.

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1600-A] Postharvest/Food Technology and Process Engineering (3)

Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A (Main Hall)

[4-1600-A-05] Optimization of Process Conditions for *Batuan* [*Garcinia binucao* (Blanco) Choisy] Fruit Powder Production

*Al Kaxier Guzman Ancheta[1], Erlinda I. Dizon[2] (1. University of the Philippines Los Banos(Philippines), 2. University of the Philippines Los Banos (Philippines))

Keywords: batuan fruit powder, response surface methodology

The study determined the optimum process conditions to produce *batuan* fruit powder using Response Surface Methodology (RSM). The factors considered were sodium metabisulfite (SMS) concentration and drying temperature. Results revealed the significant responses based on physico-chemical (titratable acidity, total soluble solids, whiteness index) and functional (antioxidant activity, total phenolics, water absorption index, water solubility index) characteristics of the powder. However, the responses that were not significant in the model were also identified based on physico-chemical (pH, bulk density, fineness modulus) and functional (none) characteristics. Hence, the optimum drying temperature and SMS concentration were found to be 50.0°C and 106 ppm, respectively. The optimum conditions were used to produce the powder to verify the predicted physico-chemical and functional properties.

Optimization of Process Conditions for
Batuan [*Garcinia binucao* (Blanco) Choisy] Fruit Powder Production

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ABSTRACT

The study determined the optimum process conditions to produce *batuan* fruit powder using Response Surface Methodology (RSM). The factors considered were sodium metabisulfite (SMS) concentration and drying temperature. Results revealed the significant responses based physicochemical (titratable acidity, total soluble solids, whiteness index) and functional (antioxidant activity, total phenolics, water absorption index, water solubility index) characteristics of the powder. However, the responses that were not significant in the model were also identified based on physicochemical (pH, bulk density, fineness modulus) and functional (none) characteristics. Hence, the optimum drying temperature and SMS concentration were found to be 50.0°C and 106 ppm, respectively. The optimum conditions were used to produce the powder to verify the predicted physicochemical and functional properties.

Keywords: *Batuan* fruit, *Batuan* fruit powder, Response surface methodology, Sodium metabisulfite, Drying

1. INTRODUCTION

Tamarind is considered as the most popular souring agent in Philippines and is used as a base in soup dishes such as *sinigang*. However, the supply of locally available tamarind may not be able to meet the huge consumer demand due to the increase in population. Philippines is continuously importing tamarind to meet the domestic needs (Valencia, 2013a; Reyes, 2000; Mojica, 2008). Thus, an alternative souring agent should be considered.

Nowadays, the potential of conversion of underutilized crops into high value products are looked upon by researchers (Ebert, 2014; Valencia, 2013b; Florido and Cortiguerra, 2003). *Batuan* fruit is one of the indigenous crops that is popular in the southern part of Philippines as souring agent instead of tamarind. A study by Quevedo *et al.* (2013) revealed the potential of using *batuan* fruit not only as a souring agent but also one that is safe for consumption and rich in nutrients. Thus, *batuan* is indeed a promising alternative souring agent.

Even though *batuan* fruit is abundant, it is seasonal so that it is available only from April to June in Philippines. Thus, preservation of this fruit is necessary to make it available all year round and to extend its shelf-life, one of which is to turn it into powder.

There are already numerous researches about production of powders from fruits such as mango powder (Jaya and Das, 2005), date powder granules (Sablani *et al*., 2008), gac fruit aril powder by spray drying (Kha *et al*., 2010), tamarind powder by drum drying using maltodextrin and arabic gum as adjuncts (Jittanit *et al*., 2011), then comparison of qualities of tamarind powder using tray and drum dryers (Khuenpet *et al*., 2012), fiber-rich powder from dragon fruit or *pitaya* peel (Senghkhamparn *et al*., 2013), and mango kernel flour using cabinet dryer (Bawar, 2013), and spray-dried soursop powder (Chang *et al*., 2018; Chang *et al*., 2019) among others. There are also published studies about *batuan* fruits' physicochemical properties, nutritional and sensory qualities (Quevedo *et al*., 2013), organic acid profile (Quevedo *et al*., 2013), and

hydroxycitric acid content that is affected during processing (Bainto *et al*., 2018). However, there is no published research yet that is specific to *batuan* fruit powder.

The production of the powder included drying, grinding, and other interventions such as inactivation of enzymes and addition of preservatives to ensure a high quality product. Drying using hot air was done to remove most of the moisture and to produce a powder with good flowability. Sodium metabisulfite (SMS) was added as anti-browning agent. Hence, this study aimed to establish the procedure to produce powder, a high-value product, from the pulp of the *batuan* fruit. Also, the optimum SMS concentration and drying temperature were determined based on the physicochemical and functional characteristics for *batuan* fruit powder processing.

2. MATERIALS AND METHODS
2.1. Time and Place of the Study
The study was conducted from September to December 2014 at the Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Baños, Laguna, Philippines.

2.2. Source of *Batuan* Fruits
Batuan fruits were obtained from a local market in Bacolod City, Philippines and then shipped to University of the Philippines Los Baños. The fruits used in the experiment had characteristic green color, hard covering, and medium to large size (about 3.7–5.5 cm in diameter) only (Fig. 1). The sizes that were used in the study were the 3 largest diameters which are medium to large (Fig. 1b).

(a)   (b)

Figure 1. Fresh *batuan* fruits in (a) bulk and (b) relative sizes used in the study.

2.3. Preparation of Frozen *Batuan* Fruits
As soon as the fresh and immature *batuan* fruits were received, the fruits were washed initially with tap water to minimize microbial load and other adhering contaminants, then, the whole fruits were disinfected by soaking in 10 ppm hypochlorous acid (HOCl) solution for 20 seconds to further decrease or eliminate its initial microbial load. The fruits were washed again in running potable water to remove excess chlorine in the fruits. Afterwards, the fruits were packed in PE bags (about 10 kg per bag), and stored in a chest freezer at -20°C to stop biochemical and microbial degradation of the fruits. Before processing into powder, the frozen fruits were thawed in running water for about 5 min and drained well.

2.4. Pre-drying Treatments
The whole fruits were cooked using a steamer for 20 min or until the color of the peel has completely changed from green to yellowish brown (Fig. 2). Steaming was necessary to inactivate the enzymes and to soften the pulp for easier removal by the pulping machine. After steaming, the cooked fruits were immediately cooled with running water to stop further heating. Then, the steamed fruits were fed through a pulping machine to separate the seeds and recover the pulp (Fig. 3 and Fig. 4).

Figure 2. *Batuan* fruits during steaming.

Figure 3. Pulping of steamed *batuan* fruits.

Figure 4. *Batuan* pulp recovered after pulping.

The recovered pulp (Fig. 4) was separated into 3 lots where each lot was treated with a predetermined amount of 10% sodium metabisulfite (SMS). SMS was added to the pulp so that the pulp would contain 0 (control), 125 ppm, and 250 ppm concentration.

2.5. Convective Drying of *Batuan* Fruit Pulp

A cabinet dryer, available in the pilot plant of Institute of Food Science and Technology, CAFS, UPLB, was used to dry the pretreated samples. The pulp (Fig. 5a) was laid on stainless steel trays layered with polyethylene to prevent the pulp from sticking onto the trays after drying. The thickness of the pulp on the trays was set at 3 mm maximum to allow faster drying. Three different drying temperatures (50°C, 60°C, and 70°C) were employed in the study. The drying of the pulp was continued until the sample reached a moisture content of about 10.75% such that the product became brittle in texture. The dried pulp (Fig. 5c) was like thin, brittle, brown flakes (ready for grinding) that were scraped using a spatula.

(a)   (b)   (c)
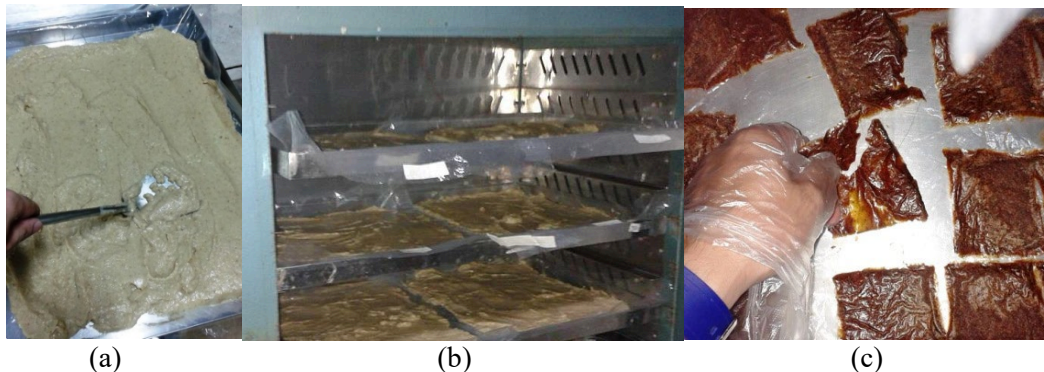
Figure 5. Drying of *batuan* pulp showing (a) spreading on trays,
(b) loading of trays inside the convective dryer, and (c) dried pulp.

2.6. Grinding and Sieving

A grinder (Koii® Platinum Edition, Koii Philippines) was used to grind the dried pulp. The produced powder was sieved using 60 mesh USA sieve to obtain a finer powder (Fig. 6b). Then the produced powder samples were immediately packed in glass bottles at room temperature for storage.

(a)   (b)
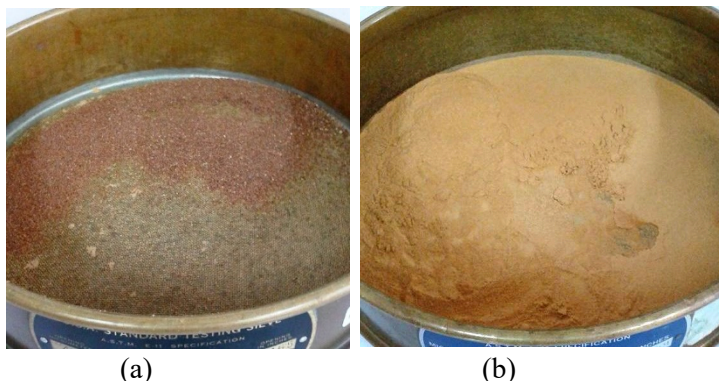
Figure 6. Sieving of *batuan* powder using 60 mesh USA sieve showing
(a) reject oversize and (b) product undersize.

2.7. Optimization of *Batuan* Fruit Powder Production

The combination of the varying SMS concentrations (0, 125 ppm, and 250 ppm) and drying temperatures (50°C, 60°C, and 70°C) resulted in 9 different treatments under study.

Based on the results of physicochemical and functional analyses, an optimum condition, which is the most acceptable, was determined using Design Expert® computer software. Statistical analysis was also done to determine whether or not the differences in the attributes are significant.

After determining the optimum condition based on analysis of physicochemical and functional properties, the optimized sample was produced following the computed optimum conditions, and analyzed of its physicochemical and functional properties to verify the correctness of the optimum condition determined using the software.

2.8. Physicochemical Analysis

The *batuan* fruit powder samples were subjected to physicochemical analysis in terms of bulk density (BD), fineness modulus (FM), pH, TA, TSS, and WI.

2.8.1. Bulk density (BD). The powder sample was filled into a pre-weighed 50-mL graduated cylinder up to the 50-mL mark. No tapping or compression of the powder was done to avoid variation in the results. The values of BD were expressed in terms of g/mL.

$$\text{Bulk Density} = \frac{\text{mass of (sample + cylinder)} - \text{mass of cylinder}}{\text{volume of sample}}$$

2.8.2. pH. The pH of the *batuan* fruit powder was determined by using a pH pen (Eutech® Instruments pH 2700, Eutech Instruments Pte. Ltd., Singapore) in a 1:9 by weight (dilution factor of 10) mixture of *batuan* powder and distilled water.

2.8.3. Titratable acidity (TA). The *batuan* fruit powder was dissolved in freshly boiled and cooled distilled water at a ratio of 1:9 by mass (dilution factor of 10). The resulting solution was added with 2-3 drops of 1% phenolphthalein indicator and titrated using 0.1 M NaOH solution up to faint pink endpoint. Then the %TA (g citric acid/100 g sample) was calculated using the formula:

$$\%\text{TA} = \frac{\text{vol. titrant used (mL)} \times \text{N titrant} \times \text{eq. wt. acid } (= 64.04 \text{ for citric acid}) \times \text{DF}}{\text{vol. sample (mL)} \times 10}$$

2.8.4. Total soluble solids (TSS). The *batuan* fruit powder was dissolved in distilled water at a ratio of 1:9 by mass (dilution factor of 10). Using a refractometer (Cole-Parmer® Refractometer EW-81150-32, Cole-Parmer Instrument Company LLC, USA), the degree Brix (°Bx) of the solution was read.

2.8.5. Whiteness index (WI). The color of the sample was measured using a chromameter (X-Rite Capsure® RM200-PT01, X-Rite Inc., USA) as *L* (lightness), *a* (redness), and *b* (yellowness). The values of *L*, *a*, and *b* obtained were used to calculate the WI using the equation according to Sheen (1990), Tsai (1994), Hsu *et al.* (2003), and Bawar (2013):

$$\text{WI} = 100 - \sqrt{(100 - L)^2 + a^2 + b^2}$$

2.9. Functional Analysis

The powder samples were also subjected to functional analysis in terms of antioxidant activity, total phenolics, WAI, and WSI.

2.9.1. Antioxidant activity. The antioxidant activity of the *batuan* fruit powder was determined based on its ability to scavenge the stable DPPH. Fifty milligrams (50 mg) of the sample was placed in a test tube and then added with 5 mL of 80% methanol solution and mixed in a vortex mixer for 10 min. The mixture was then filtered in a test tube and kept refrigerated until use. A 1-mL aliquot was obtained and added with 4 mL distilled water. Freshly prepared 1 mL of 1 mM methanolic DPPH solution was added. The solutions were allowed to stand for 30 min. The absorbance of the solutions (sample and blank) was read at 517 nm (Bawar, 2013; Murthy *et al.,* 2002 as cited in Veigas *et al.,* 2007). The percentage scavenging activity of DPPH was computed using the equation:

$$\%\text{DPPH scavenging activity} = \left(1 - \frac{\text{absorbance test sample}}{\text{absorbance blank sample}}\right) \times 100$$

2.9.2. Total phenolics content. Folin-Ciocalteau Method was used to analyze total phenolics. The powder samples were diluted to 1000 µg/mL with 80% methanol solution and then filtered using Whatman® No. 1 filter paper to remove suspended solids (that may interfere during reading of absorbance using a spectrophotometer). Exactly 0.25 mL, each, of diluted samples and standard solutions (0, 40, 80, 100, 150 µg/mL gallic acid) was obtained and diluted with 3.5 mL distilled water. Then 0.5 mL of 50% Folin-Ciocalteau reagent was added followed by 1 mL of 20% Na_2CO_3 after 3 min. Then the samples were mixed and incubated in boiling water for 1 min to develop the blue color. Then absorbance of the samples was read at 685 nm. The total phenolics content of the samples was calculated based on linear regression analysis of the standard solutions.

2.9.3. Water absorption index (WAI) and water solubility index (WSI). WAI and WSI were determined in triplicates following the method by Anderson (1982 as cited in Narbutaite *et al.,* 2008). About 1 g of each sample was suspended in 6 mL of distilled water and stirred for 30 min at 30°C. Then, the mixture was centrifuged at 4000×*g* for 20 min. The supernatant liquid was poured into a dry 15-mL test tube and stored overnight at 110°C to evaporate the water. The WAI and WSI were computed using following equations:

$$\%\text{WSI} = \frac{\text{mass of dissolved solids in supernatant}}{\text{mass of sample}} \times 100$$

$$\text{WAI} = \frac{\text{mass of sediment}}{\text{mass of sample}}$$

2.10. Statistical Analysis

All analyses were done in triplicates. The determined values were expressed as mean ± standard deviation. Data were analyzed using Analysis of Variance (ANOVA) to determine if the samples

significantly differed from one another, followed by Tukey's Honest Significant Difference (HSD) Test to know which among the samples were significantly different.

The SMS concentration and drying temperature was optimized by response surface methodology (RSM) following the procedure of Design-Expert® (Version 9.0.3.1, Stat-Ease, Inc., MN, USA) computer software. ANOVA and HSD were done using Statistical Analysis System (SAS Version 9, SAS Institute Inc., USA) computer software.

2.11. Proximate Analysis

Proximate analysis was done to further identify the best (optimized) treatment for the *batuan* powder. The procedure based on AOAC (2000) was followed.

3. RESULTS AND DISCUSSION

The effects of varying SMS concentration (0, 125 ppm, 250 ppm) and drying temperature (50°C, 60°C, and 70°C) on the quality of *batuan* fruit powder (Fig. 7) were evaluated.
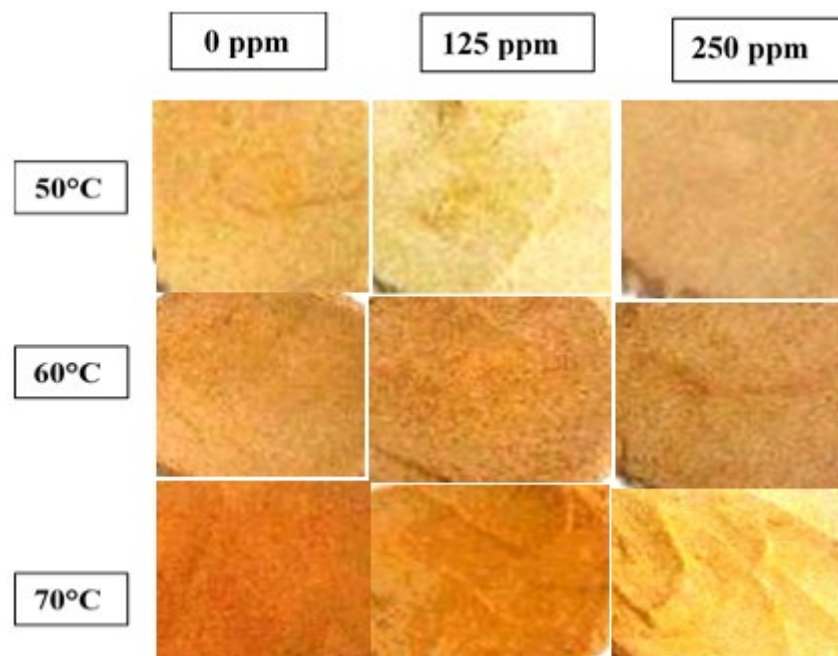
Figure 7. *Batuan* fruit powder produced at varying temperature and SMS concentration.

3.1. Effect of SMS Concentration and Drying Temperature on the Physicochemical Properties of *Batuan* Fruit Powder

3.1.1. Bulk density (BD). The bulk density accounts for the true volume occupied by the product and the volume of the voids or spaces between the particles. It is important because the volume of the packaging material necessary to accommodate a certain mass of product depends on it. Higher bulk density is favorable because at a certain mass of product, less volume is occupied and therefore the transport of the product from one place to another is easier.

The values of bulk density of *batuan* fruit powder were determined at varying temperature and SMS concentration (Table 1 and Fig. 8).

Table 1. Bulk density (g/mL) of *batuan* fruit powder at varying SMS concentration and drying temperature.

SMS CONCENTRATION (ppm)	DRYING TEMPERATURE (°C)		
	50	60	70
0	0.666 ± 0.025[a]	0.620 ± 0.005[a]	0.648 ± 0.007[a]
125	0.654 ± 0.034[a]	0.500 ± 0.002[b]	0.637 ± 0.008[a]
250	0.625 ± 0.030[a]	0.416 ± 0.003[c]	0.613 ± 0.004[a]

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.
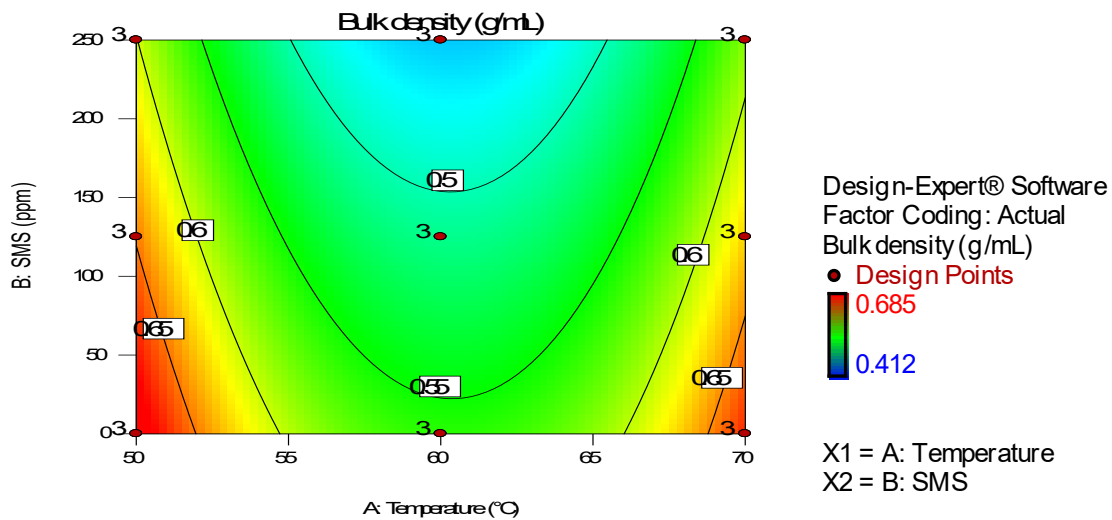
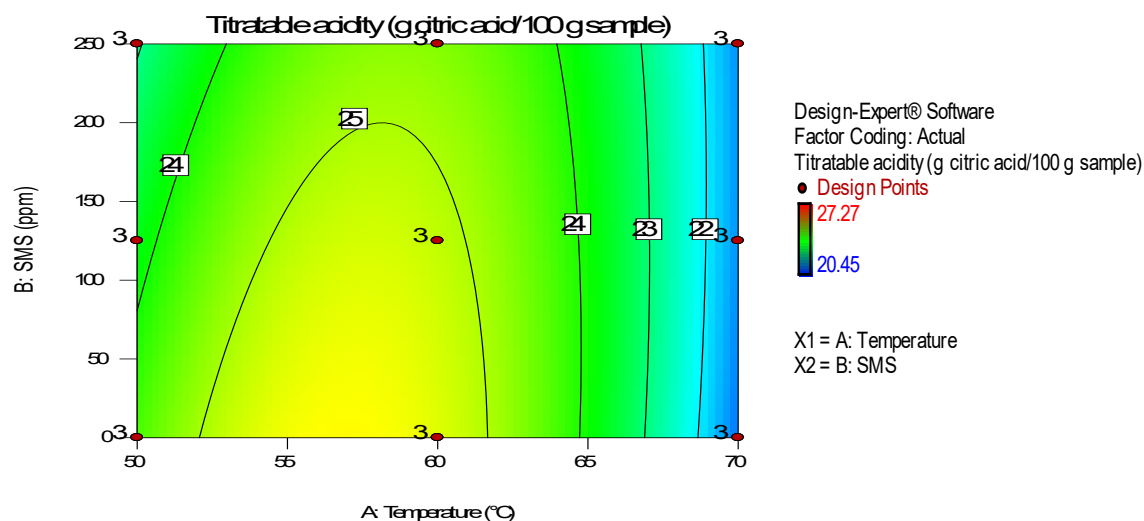Figure 8. Contour plots of bulk density of *batuan* fruit powder versus drying temperature and SMS concentration.

The BD tends to decrease somewhere between 50°C and 70°C. There is also a positive correlation between the BD and the SMS content. Increasing the drying temperature may cause further shrinkage of the pulp during drying as the volatile substances escaped from the pulp so that the resulting powder would be more compact. The decrease in bulk density at greater temperature was observed by Kha *et al.* (2010) in spray drying of gac fruit aril powder. On the other hand, higher amount of SMS may result in better protection of the pulp from oxidation where the pulp would maintain its integrity or structure, but the effect of SMS on the BD became pronounced only at 60°C (Table 1). However, there is little or no research that could verify the role of SMS in changing the BD of a powder.

3.1.2. pH. The pH indicates the acidity of the product, however, it is not a direct measure of acidity. Nevertheless, the pH is important for the *batuan* fruit powder since it is the most important property for a souring agent. The pH may determine whether or not microorganisms would survive in the sample, or what group of microorganisms may grow. Table 2 and Figure 9 show the pH values of *batuan* fruit powder at varying SMS concentrations and temperatures.

Table 2. pH values of *batuan* fruit powder at varying SMS concentration and drying temperature.

SMS CONCENTRATION (ppm)	TEMPERATURE (°C)		
	50	60	70
0	3.40 ± 0.00^a	3.33 ± 0.06^a	3.40 ± 0.00^a
125	3.40 ± 0.00^a	3.37 ± 0.06^a	3.40 ± 0.00^a
250	3.40 ± 0.00^a	3.37 ± 0.06^a	3.37 ± 0.06^a

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.
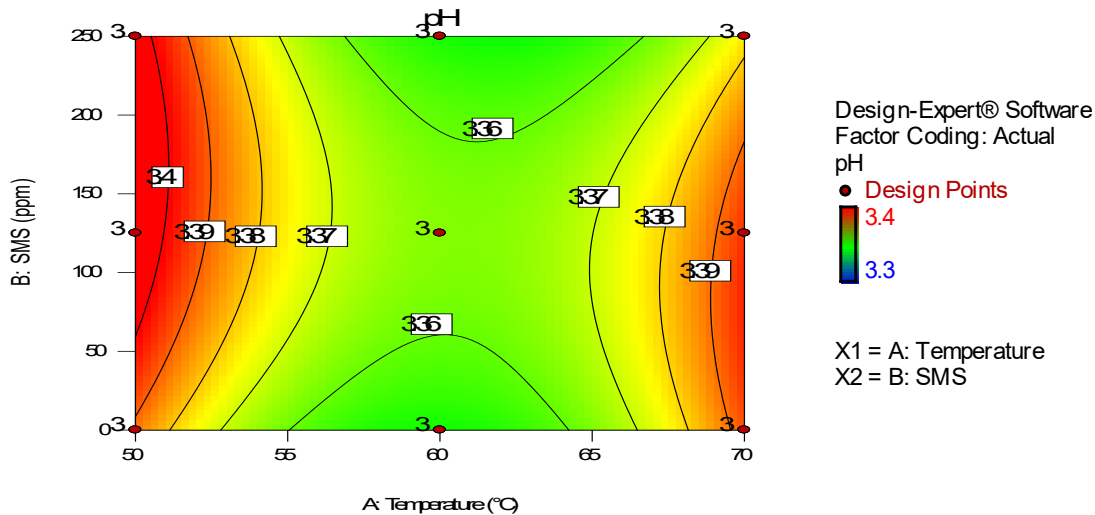
Figure 9. Contour plots of pH of *batuan* fruit powder versus
drying temperature and SMS concentration.

The differences between pH values at varying SMS concentration and drying temperature are not significant based on Two-factor ANOVA at 5% level of significance. Nevertheless, slightly lower pH values were seen at 60°C.

3.1.3. Titratable acidity (TA). The TA is a better measure of the acidity than pH. Lower pH may measure the concentration of hydrogen ions (and consequently the amount of dissociated acids) but TA accounts all the acids present whether dissociated or not. High TA values were observed in Table 3 and Figure 10. The TA was expressed as percent citric acid because it is the predominant acid present in the *batuan* fruit (Quevedo *et al.* 2017).

Table 3. Titratable acidity (g citric acid/100 g sample) of *batuan* fruit powder at varying SMS concentration and drying temperature.

SMS CONCENTRATION (ppm)	TEMPERATURE (°C)		
	50	60	70
0	24.38 ± 1.29^a	25.62 ± 1.43^a	20.87 ± 0.36^b
125	23.35 ± 0.72^a	25.41 ± 1.24^a	21.49 ± 0.95^b
250	23.35 ± 1.79^a	24.17 ± 1.24^a	21.49 ± 0.95^b

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.

Figure 10. Contour plots of titratable acidity of *batuan* fruit powder versus drying temperature and SMS concentration.

The changes in TA may be primarily due to temperature only. Moreover, at 60°C, highest values of TA were seen. However, statistical analysis at 5% level of significance revealed that there are no significant differences between the TA values at 50°C and 60°C. Varying the SMS concentration did not have significant effect on the TA due to volatilization of SMS at high temperature during drying. Nonetheless, an optimum TA may be determined somewhere between 50°C and 70°C.

3.1.4. Total soluble solids (TSS). In Table 4 and Figure 11, the TSS values are tabulated and plotted, respectively.

Table 4. Total soluble solids (degree Brix) of *batuan* fruit powder at varying SMS concentration and drying temperature.

SMS CONCENTRATION (ppm)	DRYING TEMPERATURE (°C)		
	50	**60**	**70**
0	4.60 ± 0.00^a	4.67 ± 0.12^a	5.00 ± 0.00^b
125	4.60 ± 0.00^a	4.73 ± 0.12^a	5.00 ± 0.00^b
250	4.87 ± 0.12^a	4.53 ± 0.12^a	5.07 ± 0.12^b

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.
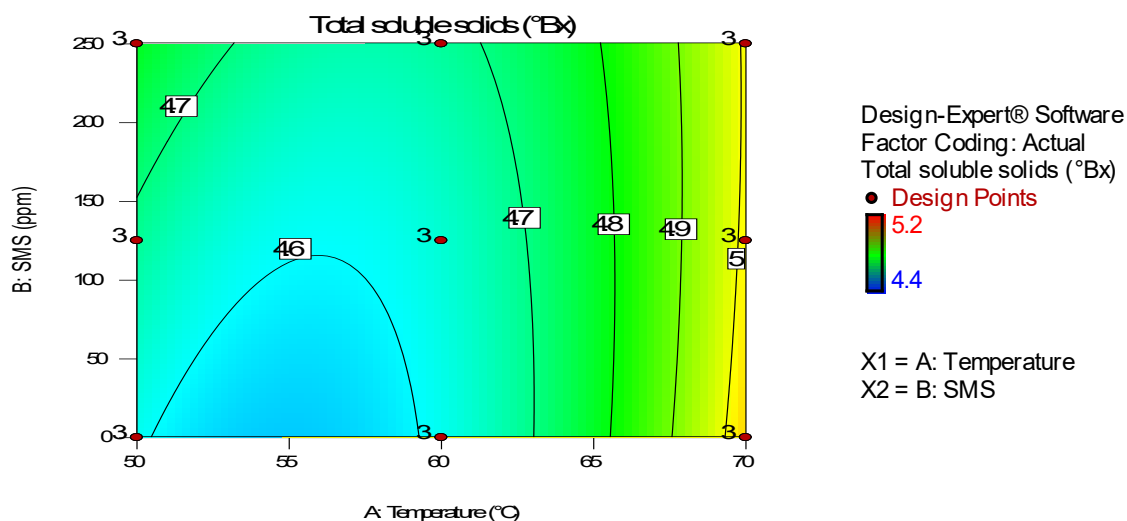
Figure 11. Contour plots of total soluble solids of *batuan* fruit powder versus drying temperature and SMS concentration.

It may be observed that the TSS generally increases with temperature, although the change in TSS was significant only at 70°C, possibly due to degradation of some components such as pectin and dietary fiber to form smaller units that are more water-soluble which were observed by Garau *et al.* (2007) in orange fruit, and de Roeck *et al.* (2008) in carrot tissue. On the other hand, the SMS did not play a role in varying the TSS of the powder. Based on Figure 11, the TSS exhibits a minimum value somewhere at 55°C.

3.1.5. Whiteness Index (WI). The values of WI range from 0 to 100 such that lighter samples have WI values approaching 100. This signifies that samples with higher WI have lighter color. The suggestion by Ajaykumar *et al.* (2012) to blanch and add sulfite to the samples was followed, and therefore, the decrease in browning (in terms of WI) of the samples was expected with increasing concentrations of SMS which is an anti-browning agent. The change in WI after varying the temperature and SMS content was shown (Table 5 and Fig. 12).

Table 5. Whiteness index values of *batuan* fruit powder at varying SMS concentration and drying temperature.

SMS CONCENTRATION (ppm)	DRYING TEMPERATURE (°C)		
	50	**60**	**70**
0	44.59 ± 2.03^{a}	44.61 ± 2.06^{a}	36.81 ± 4.55^{b}
125	46.94 ± 0.00^{a}	44.32 ± 1.02^{a}	41.34 ± 0.10^{b}
250	46.94 ± 0.00^{a}	43.85 ± 1.94^{a}	39.33 ± 3.58^{b}

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.
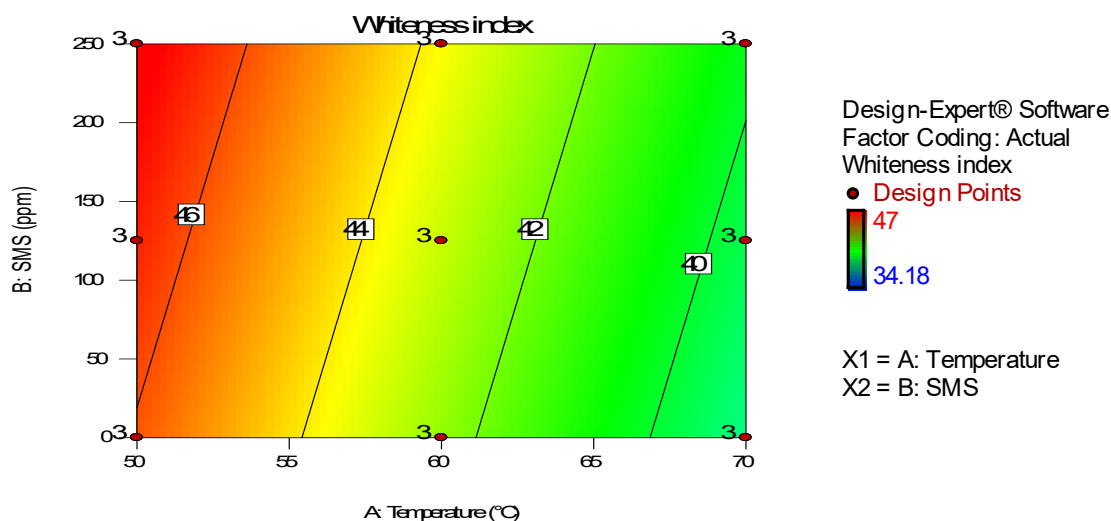
Figure 12. Contour plots of whiteness index of *batuan* fruit powder versus drying temperature and SMS concentration.

The general trend is that the WI decreased when the drying temperature increased while the WI increased slightly when SMS concentration became greater. However, based on statistical analysis at $P \leq 0.05$, even though the model is significant and the lack of fit is not significant, only the drying temperature is a significant factor that affects the WI. Nevertheless, the combined effects of temperature and SMS concentration may result in an optimum WI somewhere at the upper left corner of the region in Figure 12. The decrease in WI at increasing temperature may be due to increased rate of browning reactions at higher temperature. On the other hand, increasing the SMS content did not significantly affect the color even with the ability of SMS to inhibit browning reactions due to volatilization of SMS during drying.

3.2. Effect of SMS Concentration and Drying Temperature on the Functional Properties of *Batuan* Fruit Powder

3.2.1. Antioxidant activity. The antioxidant activity measures the ability of the *batuan* fruit powder to scavenge the stable radical DPPH. It is important to determine the antioxidant activity of the product because it is desired to know whether the product can not only satisfy the taste buds of the consumers or provide nutrients but also protect from free radicals to reduce the risk of heart disease and certain cancers. The antioxidant activity values of the samples are compared at varying SMS concentration and drying temperature (Table 6). In order to determine whether these values are competitive, the antioxidant activity values of standards 1 mM BHA and 1 mM ascorbic acid were also determined. Results show that 1 mM BHA and 1 mM ascorbic acid have DPPH-scavenging activities which are lower than the samples of *batuan* fruit powder. Hence, it may be said that the powder has a relatively high antioxidant activity. Statistical analysis shows that only the temperature, not the SMS concentration, had a significant effect on the antioxidant activity of the *batuan* fruit powder.

Table 6. Antioxidant activity (% DPPH scavenging activity) of *batuan* fruit powder at varying SMS concentration and drying temperature.

SMS CONCENTRATION (ppm)	DRYING TEMPERATURE (°C)		
	50	**60**	**70**
0	26.04 ± 0.80^a	23.99 ± 3.21^b	21.66 ± 0.56^c
125	25.05 ± 1.74^a	27.67 ± 1.72^b	19.53 ± 1.29^c
250	30.29 ± 1.07^a	21.30 ± 2.33^b	19.04 ± 0.86^c
Standard			
1 mM BHA		10.90 ± 1.93^d	
1 mM ascorbic acid		15.43 ± 3.57^e	

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.

Figure 13 shows the contour plots of antioxidant activity against SMS concentration and drying temperature. Higher antioxidant activity values are achieved at lower temperature and higher SMS content. This is because the antioxidants present in the powder, such as phenolics and ascorbic acid, are basically heat sensitive so that the antioxidants are easily degraded at higher temperature. A study by Ahmed *et al.* (2010) reported that the SMS was able to protect the phenolics and vitamin C in sweet potato flour such that the samples treated with SMS had higher total phenolics and vitamin C content than the control (without SMS). Also, increasing the drying temperature from 50°C to 60°C resulted in decreasing concentrations of total phenolics and vitamin C. However, both SMS concentration and temperature did not affect the β–carotene content of the sweet potato flour. Sulfite plays an important role as bacteriostat, antiseptic, and antioxidant. It also protects vitamin C present which may contribute to the fruit's antioxidant activity (Morgan and Field, 1929).
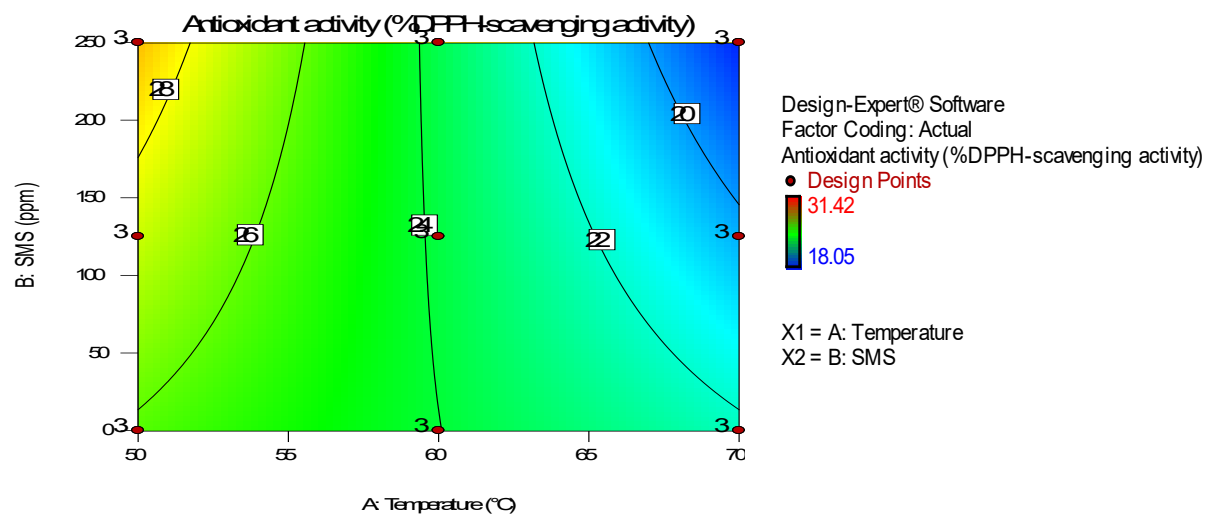
Figure 13. Contour plots of antioxidant activity of *batuan* fruit powder versus drying temperature and SMS concentration.

3.2.2. Total phenolics. The total phenolics content is usually related to the antioxidant activity of a product. In this study, gallic acid was used as a standard. The values are shown in Table 7 and Figure 14.

Table 7. Total phenolics content (µg gallic acid equivalent/mg powder) of *batuan* fruit powder at varying SMS concentrations and drying temperatures.

SMS CONCENTRATION (ppm)	DRYING TEMPERATURE (°C)		
	50	60	70
0	11.93 ± 2.65[a]	9.82 ± 2.50[a]	8.24 ± 3.33[b]
125	14.36 ± 1.13[a]	22.18 ± 3.68[a]	5.34 ± 0.83[b]
250	12.35 ± 0.98[a]	12.89 ± 0.95[a]	7.61 ± 0.80[b]

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.
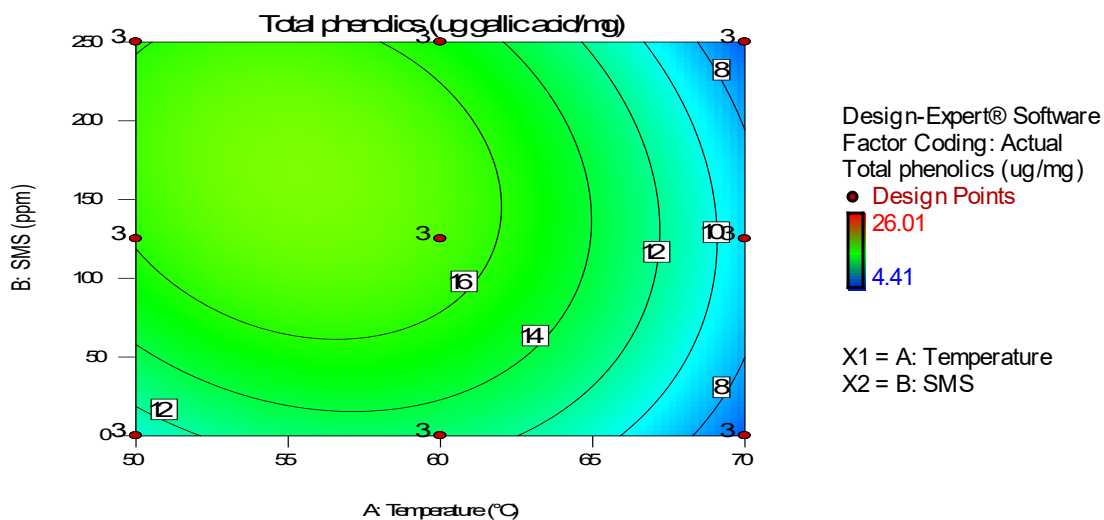
Figure 14. Contour plots of total phenolics of *batuan* fruit powder versus drying temperature and SMS concentration.

Results revealed that only the drying temperature had a significant contribution on the total phenolics content of *batuan* fruit powder. However, with combined effects of temperature and SMS concentration (Fig. 14), it is possible to identify the temperature and SMS concentration wherein a maximum total phenolics content may be predicted. Since the phenolics are heat-sensitive, it may be expected that the total phenolics was higher at lower drying temperature. The effect of SMS on the total phenolics was not significant probably because the SMS was volatile at high temperature so that the latter just escaped from the pulp during drying.

3.2.3. Water Absorption Index (WAI). WAI indicates the ability of the powder to absorb water due to the hydrophilic groups present that hold water (Narbutaite *et al.,* 2008). The WAI values are expressed as gram of sediment (equal to the sum of mass of powder and mass of water absorbed) per gram of sample. Table 8 and Figure 15 summarize the WAI values of *batuan* powder from different treatments.

Table 8. Water absorption index (g sediment/g sample) of *batuan* fruit powder at varying SMS concentration and drying temperature.

SMS CONCENTRATION (ppm)	TEMPERATURE (°C)		
	50	60	70
0	3.394 ± 0.188^a	3.617 ± 0.271^a	3.464 ± 0.227^a
125	3.449 ± 0.060^a	4.081 ± 0.018^a	3.087 ± 0.056^a
250	3.376 ± 0.156^a	4.357 ± 0.410^a	3.210 ± 0.038^a

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.
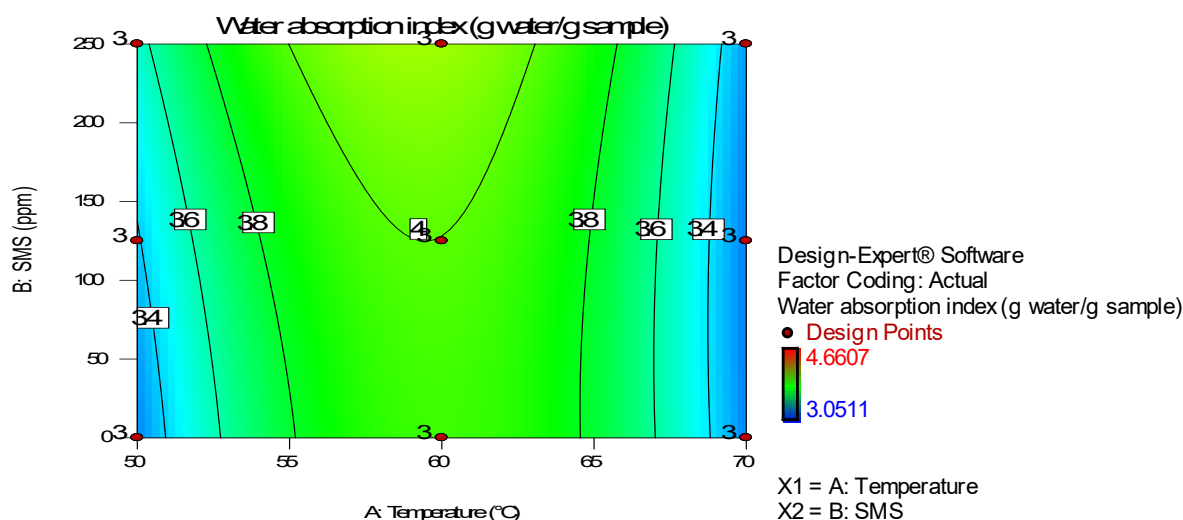
Figure 15. Contour plots of water absorption index of *batuan* fruit powder versus drying temperature and SMS concentration.

Table 8 shows that the values of WAI are not significantly different among the samples. Nevertheless, the quadratic model to express WAI as a function of SMS concentration is significant so that the model is still useful for optimization. From Figure 15, a maximum WAI may be obtained somewhere between 50°C and 70°C. The increase in WAI from 50°C to about 60°C could be due to partial gelatinization of starch and protein resulting in increased water uptake. However, above 60°C, there was higher rate of vaporization of liquids resulting in shrinkage of the polar sites and then poor absorption of moisture upon rehydration. Gunaratne and Hover (2002 as cited in Ahmed *et al.,* 2010) explained that the difference in WAI could be due to variation in the degree of engagement of hydroxyl groups to form hydrogen bonds between starch chains, and loss of starch crystalline structure.

3.2.4. Water solubility index (WSI). The water solubility index is an indication of starch degradation of the powder during drying. Table 9 and Figure 16 show the WSI values of *batuan* fruit powder at varying temperature and SMS concentration.

Table 9. Water solubility index (%) of *batuan* fruit powder at varying SMS concentration and drying temperature.

SMS CONCENTRATION (ppm)	DRYING TEMPERATURE (°C)		
	50	60	70
0	24.02 ± 1.34[a]	14.23 ± 0.41[c]	23.29 ± 1.66[a]
125	18.98 ± 0.75[b]	14.46 ± 0.37[c]	26.20 ± 1.49[a]
250	15.92 ± 1.83[b]	14.42 ± 2.37[c]	23.85 ± 1.45[a]

Mean values of the same superscript for all treatments are not significantly different at $P \leq 0.05$, HSD.
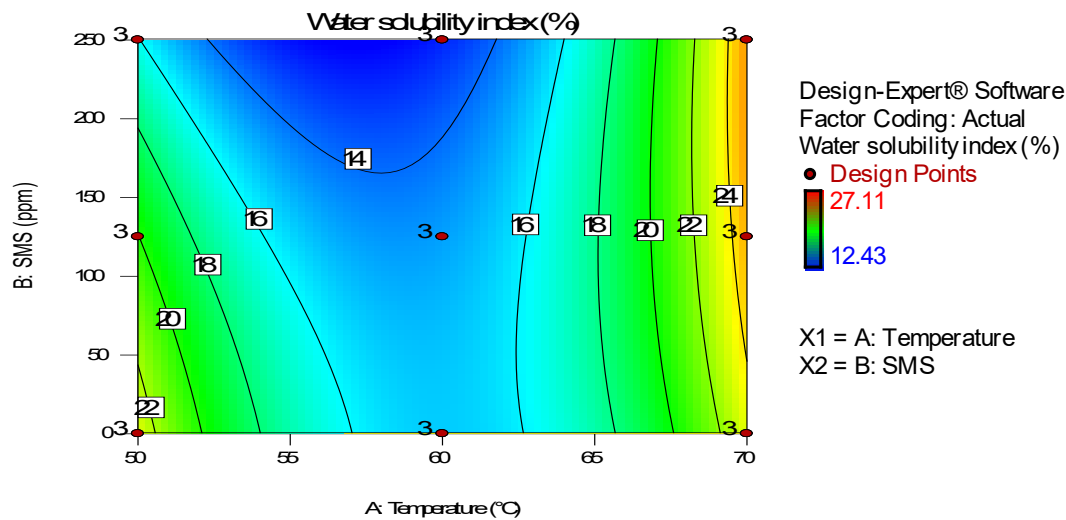
Figure 16. Contour plots of water solubility index of *batuan* fruit powder versus drying temperature and SMS concentration.

The WSI reaches a minimum value somewhere between 50°C and 70°C. Two-factor ANOVA at 5% level of significance revealed that SMS concentration and drying temperature are both significant factors that affected the WSI of the product. The model to describe the WSI of the product at varying SMS concentration and drying was also significant. However, by looking at the individual F-values, the temperature had higher value of F (32.76) than SMS concentration (8.48) which means that the temperature is the predominant factor for the variation in WSI of *batuan* fruit powder during processing.

The minimum WSI values at 60°C may be accounted to the semi-crystalline structure of starch and formation of hydrogen bonds between starch molecules. Above 60°C, the heat caused the starch molecules to swell and expose the hydrophilic groups, thereby, increasing the solubility of the powder in terms of WSI (Eliasson and Gudmundsson 1996 as cited in Ahmed *et al.* 2010). At 50°C, greater WSI values were obtained because this is where 48 hours was needed to dry the pulp. Moreover, the long drying time favored the swelling and exposure of hydrophilic groups of the starch molecules even at a slower rate.

3.3. Optimum Conditions for *Batuan* Fruit Powder

Based on the physicochemical and functional analyses, the optimum drying temperature and concentration of SMS was determined using Design-Expert® software. The physicochemical and functional properties were treated as responses and criteria for optimization. However, only those responses that have significant difference (based on Two-factor ANOVA at 5% level of significance) at varying treatments were considered. These criteria are listed in Table 10.

Table 10. Criteria for optimization of *batuan* fruit powder.

CRITERION	GOAL	IMPORTANCE	MODEL	RESULT OF ANOVA
Temperature (°C)	in range (50-70)		NA	
SMS concentration (ppm)	in range (0-250)		NA	
Whiteness index	Maximize	5	Linear	Significant
Total phenolics (mg gallic acid/g sample)	Maximize	3	Quadratic	Significant
Antioxidant activity (% DPPH-scavenging activity)	Maximize	3	2FI	Significant
Titatable acidity (g citric acid/100 g sample)	Maximize	5	Quadratic	Significant
Total soluble solids (% Brix)	Maximize	3	Quadratic	Significant
pH	None	NA	Quadratic	Not significant
Water absorption index (g sediment/g sample)	Maximize	3	Quadratic	Significant
Water solubility index (g solids/g sample)	Maximize	3	Quadratic	Significant
Bulk density (g/mL)	Maximize	3	Quadratic	Significant
Fineness modulus	None		NA	

NA – not applicable

The goal "in range" means that the optimum condition to be determined should be within the specified range. Otherwise, either "maximize" or "minimize" was selected. For example, "maximize" was chosen for whiteness index since maximum value implies lightest brown color. Hence, the determination of an optimum value involves selection of the best value for each criterion whether the latter is maximum or minimum.

The importance of a criterion was described in terms of a rating from 1 to 5 (where "5" is of highest importance). The rating of 5 was given to whiteness index because the color is the first thing that is evaluated by the consumers and hence a very important criterion. Also, since the objective of developing the product is to become an alternative souring agent, the TA was also given a rating of 5. Other factors were given 3 because it was also desired to create a product that is health beneficial (i.e., antioxidant activity, total phenolics), water soluble (i.e., water solubility index), and less spacious (i.e., bulk density).

The selection of a fit model (i.e., linear, quadratic), for the data points for each criterion, was done following the suggestion of the software. The usual options are linear, quadratic, cubic, and 2FI and the appropriate model depends on statistical analysis done by the software.

Thus, using Design-Expert® software, the optimum temperature and SMS concentration were calculated as 50.0°C and 106 ppm, respectively. Also, responses (physicochemical and functional properties) corresponding to the optimum condition were predicted by the software. However, the desirability was found to be 0.578 which is relatively low. The desirability of a response or the optimum value ranges from 0 to 1 where 0 stands for a non-acceptable value of the response and 1 where higher or lower (depending on the direction of the optimization whether maximize or minimize) values of the response have little merit (http://www.inside-r.org/packages/cran/qualityTools/docs/desirability).

The desirability of 0.578 was obtained as the geometric mean of all the individual desirability values for each response. If the goal for a certain response is to maximize its value (i.e., whiteness index), then the individual desirability approaches 1 if the obtained response is higher. Otherwise, if the goal of a response is to minimize its value (which did not happen here), then the desirability increases if the response decreases. On the other hand, if the goal is to have a target value of the response, then the desirability approaches 1 if the response is nearer to the target value

(http://www.jmp.com/support/help/Desirability_Profiling_and_Optimization.shtml). However, since the optimization of SMS concentration and drying temperature involved 8 responses (only those that were significantly affected by SMS and temperature based on ANOVA) with different goals, then the resulting optimum value, together with the desirability, would lie midway to satisfy all the responses (Kuhn, 2012). Furthermore, the desirability may score how the predicted values agree with the desired (maximum or minimum) values. For example, for whiteness index (WI), the maximum possible value that was obtained was 46.94 at 50°C and 250 ppm SMS, however, the predicted value of WI at the optimum SMS concentration and drying temperature is 46.4773 (Table 13) which is slightly lower than the maximum and may give an individual desirability near 1. On the other hand, for water absorption index, where the goal is to maximize its value, the maximum possible value is 4.357 at 60°C and 250 ppm SMS, however, the predicted value is only 3.36619 which is way below the desired value resulting in a low individual desirability. Therefore, the overall desirability of 0.578 was a result of high and low values of individual desirability. Nevertheless, since the color (in terms of WI) is one of the most important responses during optimization (where a score of 5 was given for its importance), it was observed that its predicted value was very near to the desired value. However, because the other responses were given less importance, the individual desirability for the other responses became low which consequently gave a lower overall desirability.

Nonetheless, determining the actual responses (by producing the *batuan* powder at the suggested optimum SMS concentration and drying temperature) is more important than just predicting the responses and comparing them with the desired responses.

3.4. Adoption of Predicted Optimum Conditions for *Batuan* Fruit Powder

Even with relatively low desirability, the computed optimum value was followed to produce optimized *batuan* fruit powder. The drying of *batuan* pulp (containing 106 ppm SMS) took 48 hours at 50°C (dryer could not attain the exact optimum temperature of 50.0°C) to produce the brittle dried pulp ready for grinding. After the production of the optimized product, the powder was tested for its physicochemical and functional properties to verify the predicted values as shown in Table 11.

Table 11. Predicted versus observed properties of optimized *batuan* fruit powder
(50.0°C drying temperature, 106 ppm SMS).

PHYSICOCHEMICAL/ FUNCTIONAL PROPERTY	PREDICTED VALUE	ACTUAL VALUE	ERROR (%)
Whiteness index	46.4773	48.30 ± 1.51	3.77
Total phenolics (mg gallic acid/g sample)	15.6955	31.13 ± 0.90	49.58
Antioxidant activity (% DPPH-scavenging activity)	27.1375	26.04 ± 0.80	4.21
Titratable acidity (g citric acid/100 g sample)	23.8629	26.56 ± 0.61	10.15
Total soluble solids (% Brix)	4.66923	4.9 ± 0.1	4.71
pH	NA	1.58 ± 0.07	NA
Water absorption index (g sediment/g sample)	3.36619	3.0094 ± 0.1148	11.86
Water solubility index (g solids/g sample)	20.5724	21.837 ± 5.647	5.79
Bulk density (g/mL)	0.654995	0.661 ± 0.001	0.91
Fineness modulus	NA	0.70 ± 0.06	NA

NA – not applicable

Table 11 suggests that some of the observed values agree with the predicted values, such as whiteness index, antioxidant activity, TSS, WSI, and bulk density, with small percent error. On the other hand, large

percentage of error was observed in values for total phenolics. This may be a result of lack of fitness of the models during optimization. For each response, an appropriate model (i.e., linear, quadratic, cubic, 2FI), which was also suggested by the software, was needed to be established to describe its variation with SMS concentration and drying temperature. However, based on statistical analysis at 5% level of significance, except for WI and TA, the lack of fitness of most of the responses were found to be significant, meaning, the models could not satisfactorily predict the values of these responses at a given SMS concentration and drying temperature. Even so, the models were still found to be useful since they are significant based on statistical analysis and they are necessary for optimization. Nonetheless, the actual values of most of the responses (i.e., WI, total phenolics, TA, WSI, BD) were found to be better than the predicted values.

3.5. Comparison of Optimized *Batuan* Fruit Powder Against *Batuan* Fruit Pulp

To describe the drying characteristics of the optimized *batuan* fruit powder, the physicochemical and functional properties of the latter were compared with those of the wet pulp obtained after steaming and pulping (Table 12).

Table 12. Proximate composition of *batuan* fruit pulp and powder.

COMPOSITION	WET BASIS (%)		DRY BASIS (g water/g solids)	
	PULP	POWDER	PULP	POWDER
Moisture content	87.15 ± 0.10	10.75 ± 0.02	6.7849^a	0.1205^b
Ash	0.32 ± 0.02	2.44 ± 0.06	0.0251^a	0.0273^a
Crude protein	2.81 ± 0.07	0.23 ± 0.05	0.2189^a	0.0026^b
Crude fat	1.84 ± 0.52	3.16 ± 0.11	0.1435^a	0.0354^b
Crude fiber	0.45 ± 0.03	8.23 ± 0.11	0.0922^a	0.0351^b
NFE	7.42	75.19	0.5775	0.8425

Mean values with the same superscript within rows are not significantly different at $P \leq 0.05$, HSD.

Results show that there was no significant decrease in total ash content. However, moisture content, crude protein, crude fat, and crude fiber decrease significantly after drying.

Drying process involves the removal of water through evaporation to decrease the moisture content of food and consequently to lengthen its shelf-life. This is the main reason for the significant decrease in moisture content of the pulp samples from 87.15% to 10.75%. Decrease in crude protein content of *batuan* powder is attributed to Maillard browning that might have occurred in the sample (Saltmarch and Labuza, 1982). Proteins were used up by non-enzymatic browning by reacting with a reducing sugar to form brown compounds (melanoidins). On the other hand, the decrease in crude fat content can be attributed to lipid oxidation in powdered sample, however, the rancidity may not be pronounced due to several factors such as minute amount of crude fat in the sample, limited oxygen due to the impermeability of the packaging material to gases, and the presence of antioxidants that prevent lipid oxidation. The very low water activity of the powder beyond the monolayer value could have favored the occurrence of lipid oxidation in the powdered sample since water would not have a "protective effect" to prevent lipid oxidation, thus, lowering the crude fat content of the sample. Significant decrease in crude fiber may be credited to two main reasons: (1) curling and twisting of fibers during drying making it more prone to degradation, and (2) during sieving of pulverized sample with 60 mesh screen, incorporation of fibrous components in the reject oversize.

Aside from the proximate composition, some of the functional properties of wet pulp and powder were also compared as seen in Table 13.

Table 13. Functional properties of *batuan* fruit pulp and powder.

COMPOSITION	WET BASIS (%)		DRY BASIS (g water/g solids)	
	PULP	POWDER	PULP	POWDER
Antioxidant activity (% DPPH-scavenging activity)	27.60 ± 3.40[a]	26.04 ± 0.80[a]	NA	NA
Total phenolics (mg GAE/g sample)	5.58 ± 0.79	31.13 ± 0.90	0.434[a]	0.3488[b]

Mean values with the same superscript within rows are not significantly different at $P \leq 0.05$, HSD.
GAE – gallic acid equivalent

Table 13 shows that the antioxidant activity of the *batuan* fruit pulp remained unchanged even after drying and grinding. The total phenolics content (in wet basis), on the other hand, was found to increase after drying. However, if the total phenolics of pulp and powder are to be converted to dry basis (mg GAE/g solids), the resulting values are 0.434 and 0.3488, respectively, meaning, the amount of phenolics actually decreased during drying. The phenolics serve as antioxidants that protect the product during drying where oxidation could happen. However, for *batuan* fruit pulp, the antioxidant activity remained unchanged so that the phenolics did not play the major role as antioxidants. There may be other compounds present in the *batuan* that contributed largely to the antioxidant activity such as carotenoids which are more heat-stable than phenolics and ascorbic acid. Studies by Zhang and Hamauzu (2004) and Turkmen *et al.* (2005) show that phenolics and ascorbic acid were reduced during conventional heat treatment of selected vegetables, but the carotenoids were retained after cooking. A study by Torres *et al.* (2014) revealed that the carotenoids, although known to be nonpolar, in seaweeds, is highly soluble in methanol. The carotenoid present in the seaweeds is primarily zeaxanthin, a carotenoid alcohol that is heat stable, which is a powerful antioxidant in fruits and vegetables. This implies that the *batuan* fruit may contain significant amount of zeaxanthin that primarily affects the antioxidant activity of the fruit, however, detection and quantification of such compound is beyond the scope of this study.

4. CONCLUSION

During the production of *batuan* fruit powder, the procedure was established and the process conditions were optimized. The drying temperature was set at 50°C, 60°C, and 70°C, while the concentration of SMS in the wet pulp was varied at 0, 125 ppm, and 250 ppm. The optimum temperature and SMS concentration were determined using Design-Expert® software where the physicochemical and functional characteristics of the powder were considered as the responses. Hence, the optimum temperature and SMS concentration were found to be 50.0°C and 106 ppm, respectively, with a desirability of 0.578. The optimum conditions were used to produce the powder, and the powder was evaluated again of its physicochemical and functional properties.

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5:15 PM - 5:30 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A)

[4-1600-A-06] Effect of Processing Conditions on Bioactive Compounds and Antioxidant Activities of Tea Infusion

*Wei Qin[1], Sunantha Ketnawa[1], Florencio, Jr. Collado Reginio[1,2], Ryutaro Yamada[3], Takuya Araki[3], Yukiharu Ogawa[1]　(1. Graduate School of Horticulture, Chiba University.(Japan), 2. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Baños(Philippines), 3. Institute of Fruit Tree and Tea Science, NARO(Japan)）
Keywords: Antioxidant activities, Bioactive compounds, Coarse kneading, Japanese green tea, Tea processing

The manufacturing process of Japanese green tea consists of six steps, which are followed by steaming, coarse kneading, crumpling, secondary kneading, precise kneading and drying. Coarse kneading (CK) is the first step of processing to destroy the structure of tea leaf at a certain temperature. Previous studies have shown that coarse kneading have the greatest impact on green tea quality during tea processing. However, few studies to date have provided information regarding the effects of coarse kneading on tea quality during the Japanese green tea manufacturing process. Thus, this study aimed to investigate the effects of processing conditions in coarse kneading step on bioactive compounds and antioxidant activities in tea infusion. The first flush fresh tea leaves were harvested at the experimental field of the national agriculture and food research organization (NARO) in Shizuoka, Japan in May 2019, and were processed continuously with different CK temperatures (34, 36 and 38 ˚C). The sample tea leaves were collected at each processing step and immediately cooled in a refrigerator. The processed tea leaves at each step were infused by hot distilled water at 95 ˚C for 5 min to examine bioactive compounds (total polyphenol content (TPC), total flavonoid content (TFC), and chlorogenic acid content (CA)) and antioxidant activities (1,1-diphenyl-2-picrylhydrazyl (DPPH) and 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulphonic acid (ABTS) radical scavenging activity, ferric-reducing antioxidant power (FRAP) and metal ion chelating activity (MIC)). The results showed that bioactive compounds (TPC, TFC and CA) of tea infusion increased with increasing CK temperature. The values of the antioxidant activities of the tea infusion also showed a positive correlation to CK temperatures. ABTS and MIC trend of tea infusions showed the same trend as bioactive compounds as well as they were the highest at 38 ˚C CK. However, DPPH and FRAP values of tea infusions produced from 36 and 38 ˚C CK did not follow the same trend demonstrating higher of those value of 36 ˚C than those of 38 ˚C. In general, antioxidant activities trend of 38 ˚C tea infusion was more stable than those of 34 and 36 ˚C during the tea manufacturing process. These results indicate that coarse kneading temperature during green tea processing have a positive effect on the increase in the bioactive compounds of tea infusion and the stability of the antioxidant activities.

5:30 PM - 5:45 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A)

[4-1600-A-07] *In Vitro* Release Characteristics of Sugars and Hydrolysis of Starch During Simulated Digestion of Saba banana at Different Maturity Stages

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Keywords: maturity, starch digestibility, viscosity, sugars

Saba banana [*Musa* 'saba' *(Musa acuminata x Musa balbisiana)*] is one of the most popular fruit crops in terms of production and trade in the Philippine food industry. It has a wide range of applications in the domestic market and is also known to offer great nutritional and health benefits. However, restriction of banana consumption as desserts or snacks is suggested as it also contains higher amounts of carbohydrates compared to other fruits. The determination of bioavailable carbohydrate in a given product is necessary to predict its effect on postprandial blood glucose response. Thus, this study was conducted to examine the changes in sugar and starch fractions of Saba banana during simulated *in vitro* gastrointestinal digestion. Five maturity stages of Saba banana were identified based on peel color index (1, all green; 2, green but turning yellow; 3, greenish yellow; 4, yellow with green tips; and 5, yellow with brown flecks). Unhomogenized cut and homogenized slurry samples representing intact tissue and structure-less states of Saba banana were obtained in each maturity stage and evaluated for starch hydrolysis rates. Physicochemical properties during maturation, and release characteristics of sugars (sucrose, fructose, and glucose) and viscosity of digesta during *in vitro* digestion were also determined. The decrease in total starch content was an indication that starch degradation occurred during ripening. More than 70% of resistant starch was degraded from unripe to overripe stage. During the course of digestion, a significant decrease in sucrose was observed whereas glucose and fructose contents increased concomitantly. The highest starch hydrolysis (%) for both homogenized and unhomogenized states was determined in unripe stages and a decreasing trend was observed as maturity advanced. The higher digesta viscosity values of ripe stages than unripe counterpart could influence its susceptibility to digestion by reducing the extent of mixing and the interaction with digestive enzymes. Additionally, comparing the two states, the unripe stages of homogenized slurry samples were found to have significantly higher percent starch hydrolysis and shorter digestion time than unhomogenized cut samples owing to the difference in structure. These results provide an understanding of the different factors that significantly impact starch digestibility during maturation of Saba banana.

5:45 PM - 6:00 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A)

[4-1600-A-08] *In Vitro* Examination of Starch Digestibility and Antioxidant Activities of Amaranth Grains

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Keywords: Amaranth grain, In vitro digestion, Antioxidant activities

Amaranth grain is a pseudo-cereal crop which is rich in both macro- and micronutrients. Apart from having high starch content, the phytochemicals in amaranth grains are abundant especially phenolic acids. In this study, the digestibility of starch in amaranth was examined by means of an *in vitro* simulation and the total phenolics, flavonoids, and antioxidant activities of two different solvent extracts from raw and cooked grains were determined. Whole grain and slurry states of cooked amaranth were subjected to simulated *in vitro* gastrointestinal digestion and starch hydrolysis (%) was computed based on the free glucose content of the digested fractions collected at different time periods. Additionally, raw and cooked whole amaranth grains were evaluated for their total phenolic content (TPC), total flavonoid content (TFC) and antioxidant activities (ferric reducing antioxidant power [FRAP], DPPH and ABTS free-radical scavenging activity, and metal ion chelating [MIC] activity) using sodium phosphate buffer (100mM, pH 7.4) and 80% (v/v) methanol as extracting solvents. Results showed that after 360 min of *in vitro* gastrointestinal digestion, the whole grain cooked amaranth grain showed a relatively lower starch hydrolysis (66.82 ± 1.60%) than the slurry state

(80.44±3.91%)., This was accounted to the differences in morphological structure. It was also found that raw amaranth grain showed higher TPC, TFC, and antioxidant activity values when compared to cooked grain, except for DPPH, and these phenolic compounds were released more in sodium phosphate buffer than in methanol solvent possibly due to more water-soluble compounds of the grain. The results indicated that amaranth grain could be regarded as a healthy food as it can be a potential source of natural antioxidant and, at the same time, whole amaranth grain could help in controlling blood glucose level changes because of its slow rate of digestion.

Oral Session | Postharvest/Food Technology and Process Engineering
[4-1600-A] Postharvest/Food Technology and Process Engineering (3)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Hall A (Main Hall)

[4-1600-A-09] Effects of Cell Structure Changes of Citrus Peel on the Digestibility of Intracellular Antioxidants during *in vitro* Digestion

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Keywords: Plant foods, Bioavailability, Antioxidant activity, In vitro, Structural attributes

Plant foods are essential foods in human diets. Vegetables and fruits contain a lot of antioxidants which can reduce the risk of some chronic diseases. In most cases, food processing during industrial or household meal preparation may result in microstructural changes and significant loss of natural antioxidants. Assessing the impact of food processing on the release of natural antioxidants during digestion is a critical factor for maintaining and improving its bioavailability. In this study, relationship between structural changes of plant cell and changes in antioxidant potential during *in vitro* digestion was investigated using four particle sizes of freeze-dried citrus peel powder. These were regarded as plant cell models with various degree of cell damage. Simulated *in vitro* gastrointestinal digestion was conducted and digested fractions were taken at different digestion times. The supernatant fractions were used to evaluate the total polyphenol content [TPC] and antioxidant activity (DPPH-radical scavenging activity and ferric reducing antioxidant power [FRAP]). Results showed that the antioxidant potential of digested fractions during *in vitro* digestion was approximately decreased with decreasing powder size. Moreover, DPPH and FRAP activities of all sample in the gastric digestion stage showed higher values than that of small intestinal stage. In the contrary, TPC showed high values in the small intestinal phase of digestion. These results suggest that the release of antioxidant compounds in plant foods during digestion could be related to their structural attributes such as cell damages, which can be affected by different processing conditions.

Effects of Cell Structure Changes of Citrus Peel on the Digestibility of Intracellular Antioxidants during *in vitro* Digestion

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ABSTRACT

Plant foods are essential foods in human diets. Vegetables and fruits contain a lot of antioxidants which can reduce the risk of some chronic diseases. In most cases, food processing during industrial or household meal preparation may result in microstructural changes and significant loss of natural antioxidants. Assessing the impact of food processing on the release of natural antioxidants during digestion is a critical factor for maintaining and improving its bioavailability. In this study, relationship between structural changes of plant cell and changes in antioxidant potential during *in vitro* digestion was investigated using three particle sizes of freeze-dried citrus peel powder. These were regarded as plant cell models with various degree of cell damage. Simulated *in vitro* gastrointestinal digestion was conducted and digested fractions were taken at different digestion times. The supernatant fractions were used to evaluate the total polyphenol content [TPC] and antioxidant activity (DPPH-radical scavenging activity and ferric reducing antioxidant power [FRAP]). Results showed that the antioxidant potential of digested fractions during *in vitro* digestion was approximately decreased with decreasing powder size. Moreover, DPPH and FRAP activities of all sample in the gastric digestion stage showed higher values than that of small intestinal stage. In the contrary, TPC showed high values in the small intestinal phase of digestion. These results suggest that the release of antioxidant compounds in plant foods during digestion could be related to their structural attributes such as cell damages, which can be affected by different processing conditions.

Keywords: Plant foods, Bioavailability, Antioxidant activity, *In vitro*, Structural attributes, Cell damage

Oral Session | Postharvest/Food Technology and Process Engineering

[4-1600-C] Postharvest/Food Technology and Process Engineering (4)

Chair:Kornkanok Aryusuk(King Mongkut's University of Technology Thonburi, Thailand), Itaru Sotome(University of Tokyo, Japan)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C (3rd room)

[4-1600-C-01] **Estimation of Moisture Loss of Cucumber during Storage using CFD Simulation based on Heat and Mass Transfer Model**

*Seong-Heon Kim[1], Chinatsu Nishihara[1], Fumina Tanaka[1], Fumihiko Tanaka[1] （1. Kyushu Univ.(Japan)）

　4:00 PM - 4:15 PM

[4-1600-C-02] **Screening (*in vitro*) The Inhibition Effect of Generally Recognized As Safe (GRAS) Substances on The Postharvest Fungal Pathogens and Its Modelling**

*Passakorn Kingwascharapong[1], Supatra Karnjanapratum[2], Fumina Tanaka[1], Fumihiko Tanaka[1] （1. Kyushu University(Japan), 2. King Mongkut's Institute of Technology Ladkrabang(Thailand)）

　4:15 PM - 4:30 PM

[4-1600-C-03] **Modeling The Ripening Behavior of Mature Green Tomato at Different Storage Temperatures**

*Drupadi Ciptaningtyas[1,2], Wakana Kagoshima[3], Rei Iida[1], Hitomi Umehara[1], Masafumi Johkan[1], Takeo Shiina[1] （1. Graduate School of Horticulture, Chiba University(Japan), 2. Faculty of Agro-industrial Technology, Universitas Padjadjaran(Indonesia), 3. Faculty of Horticulture, Chiba University(Japan)）

　4:30 PM - 4:45 PM

[4-1600-C-04] **Quality and Shelf-life Prediction of Fresh-cut ‘Phulae’ Pineapple by Using Image Analysis and Artificial Neural Networks**

*Rattapon Saengrayap[1], Mayura Dongsuea[1] （1. Postharvest Technology and Logistics Program, School of Agro-Industry, Mae Fah Luang University(Thailand)）

　4:45 PM - 5:00 PM

[4-1600-C-05] **Stationary Machine Vision Based Real-Time Estimation of Japanese Black Cattle Serum Vitamin A using Eye Fundus Color**

*SAMUEL OUMA OTIENO[1], Naoshi Kondo[1], Tateshi Fujiura[1], Yuichi Ogawa[1], Tetsuhito Suzuki[1], Katsuya Takenouchi[1], Hidetsugu Yoshioka[1], Moriyuki Fukushima[2], Takahiko Ohmae[3] （1. Graduate School of Agriculture, Kyoto University(Japan), 2. Hyogo Prefectural Hokubu Agricultural Institute(Japan), 3. Tajima Agricultural High school(Japan)）

　5:00 PM - 5:15 PM

[4-1600-C-06] **Segmentation of common scab lesion on intact potatoes using single near-infrared image**

*Dimas Firmanda Al Riza[1,2], Kazuya Yamamoto[3], Kazunori Ninomiya[3], Tetsuhito Suzuki[1], Yuichi Ogawa[1], Naoshi Kondo[1] （1. Laboratory of Biosensing Engineering, Graduate School of Agriculture, Kyoto University, Kitashirakawa 6068267, Kyoto(Japan), 2. Department of Agricultural Engineering, Faculty of Agricultural Technology, University of Brawijaya, Jl. Veteran 65145, Malang(Indonesia), 3. Product Planning Department,

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Shibuya Seiki Co., Ltd. 2200, Minamiyoshida, Matsuyama, Ehime, 791-8042(Japan)）

5:15 PM - 5:30 PM

[4-1600-C-07] **Myanmar Mango Maturity Prediction Based on Skin Color Using Machine Vision System**

*RULIN CHEN[1], Dimas Firmanda Al Riza[1], Thwe Thwe Tun Naw[2], Phyu Phyu Leiyi[2], Aye Aye Thwe[2], Khin Thida Myint[1], Yuichi Ogawa[1], Tetsuhito Suzuki[1], Naoshi Kondo[1] （1. Kyoto University(Japan), 2. Yezin Agricultural University(Myanmar)）

5:30 PM - 5:45 PM

[4-1600-C-08] **Measurement of Chicken Eggshell Optical Properties Using Terahertz Spectroscopy**

*Alin Khaliduzzaman[1,3], Keiji Konagaya[1], Tetsuhito Suzuki[1], Ayuko Kashimori[2], Naoshi Kondo[1], Yuichi Ogawa[1] （1. Graduate School of Agriculture, Kyoto University(Japan), 2. NABEL Co., LTd.(Japan), 3. Department of Food Engineering and Technology, Sylhet Agricultural University(Bangladesh)）

5:45 PM - 6:00 PM

[4-1600-C-09] **Application of LCA (Life Cycle Assessment) Methodology in Bioethanol Production from Sugar Industry Wastewater (Molasses) – A Case Study in West Java Province, Indonesia**

*Agusta Samodra Putra[1,2], Ryozo Noguchi[3], Tofael Ahamed[3] （1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Research Center for Chemistry, Indonesian Institute of Sciences(Indonesia), 3. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan)）

6:00 PM - 6:15 PM

4:00 PM - 4:15 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C)

[4-1600-C-01] Estimation of Moisture Loss of Cucumber during Storage using CFD Simulation based on Heat and Mass Transfer Model

*Seong-Heon Kim[1], Chinatsu Nishihara[1], Fumina Tanaka[1], Fumihiko Tanaka[1]　（1. Kyushu Univ.(Japan)）
Keywords: Cucumber, Heat and mass transfer, Moisture content, Postharvest, Storage

Maintaining the quality of perishable products that contain high moisture is considered as one of the primary goals to extend their shelf life in the postharvest process. Cucumber is one of the popular fruits due to its unique flavor and crunchy texture, and also a perishable product. In addition, cucumber fruit is mostly consumed raw, without cooking. As the fruit consists of approximately 95% of moisture, cucumber is prone to lose easily its marketability depending on storage conditions. This study was conducted to simulate the heat and mass transfer phenomena of Japanese cucumber during storage using mathematical equations based on drying model to predict the amount of moisture releasing from the fruit, and presented a useful guideline for cucumber storage. The mathematical models were developed based on heat and mass transfer phenomena and were appropriately modified for storage condition. A 3D geometry of cucumber was reconstructed by using an X-ray CT scanner. The simulation was carried out using COMSOL software. To verify the model, the actual data, such as the changes in moisture content for 8 days under 4 different environmental conditions and temperature for 100 minutes under 2 different conditions, were investigated and then compared with the simulated data. As a result, the accuracy of the simulation of moisture ratio and temperature change was estimated at approximately 0.4% and 0.4°C in average RMSE, respectively. This study would be helpful for designing the optimal postharvest process and creasing economic profits on food storage.

4:15 PM - 4:30 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C)

[4-1600-C-02] Screening (*in vitro*) The Inhibition Effect of Generally Recognized As Safe (GRAS) Substances on The Postharvest Fungal Pathogens and Its Modelling

*Passakorn Kingwascharapong[1], Supatra Karnjanapratum[2], Fumina Tanaka[1], Fumihiko Tanaka[1]　（1. Kyushu University(Japan), 2. King Mongkut's Institute of Technology Ladkrabang(Thailand)）
Keywords: GRAS, B.cinerea, modeling

Botrytis cinerea is one of ubiquitous fungal pathogens, mainly found in several kinds of citrus fruits and stone fruits. The use of mathematical models for quantifying and predicting microbial density has gained increasing attention because it is useful to assess biological hazards in human and animal healthcare. This study aimed to screen the effect of GRAS (Generally Recognized As Safe) substances (sodium benzoate, sodium propionate and sodium dehydroacetate) on the inhibition of mycelium growth (diameter: mm) of pathogenic fungi, *Botrytris cinerea, in vitro* study, and also to model the efficacy of antifungal activity of GRAS substances by using mathematical models. The influence of GRAS substances at different concentrations (0.1-2%) was used to evaluate antifungal activity. Sodium dehydroacetate at 0.1% showed the highest effectiveness in inhibition *B. cinerea* than the other substances at 25º C during 45 days of incubation. Three mathematic models, including modified logistic model, modified Gompertz model, and Baranyi and Robert model, were employed to predict the growth curve of *B. cinerea* treated with sodium propionate, where the root mean squares error (RMSE) and R^2 were employed to evaluate the performance of each model. The modified

logistic model showed the highest performance with the satisfactory statistical indices (highest R^2 and lowest RMSE) which indicated the better fit than other models. It can be concluded that sodium dehydroacetate is a potential GRAS substance in inhibiting *B. cinerea* and modified logistic model is a useful model to evaluate the mold growth under the various concentrations of sodium propionate treatment.

4:30 PM - 4:45 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C)

[4-1600-C-03] Modeling The Ripening Behavior of Mature Green Tomato at Different Storage Temperatures

*Drupadi Ciptaningtyas[1,2], Wakana Kagoshima[3], Rei Iida[1], Hitomi Umehara[1], Masafumi Johkan[1], Takeo Shiina[1]
 （1. Graduate School of Horticulture, Chiba University(Japan), 2. Faculty of Agro-industrial Technology, Universitas Padjadjaran(Indonesia), 3. Faculty of Horticulture, Chiba University(Japan)）
Keywords: mature green stage, modeling, red color development, ripening, tomato

Mature green tomato is a tomato that is physiologically mature but pre-ripe and has a green and slightly white color in appearance. In many countries except Japan, tomatoes are harvested in the mature green stage of development for commercial purpose. Some researcher recommends the mature green stage of development for the harvesting time of typically red tomato. It was studied that harvesting tomato at the mature green stage can extend the shelf life of the tomato which is desirable in the long-distance distribution and long-term storage. The previous study has proven that even though every individual tomato that was stored at the same storage temperature has a different onset in the ripening indicator, but the trend was almost the same and it is possible to make a model of it. This study was aimed to observe the red color development of mature green tomato during ripening while the lag time of red color development for tomato stored at different storage temperature conditions was being observed. Furthermore, the standard curve for predicting the red color development based on the storage period of mature green tomato (cv. Momotaro York) at different storage temperature will also be developed. Five storage temperatures (12, 15, 20, 25, and 30 ˚C) were assigned to hold 5 tomatoes (cv. Momotaro York), respectively until every tomato reached the fully ripening stage. Red color development was determined by measuring the CIE a* value of 7 spots in the surface area of the tomato. Lag time will be determined based on the breaker stage of development of the tomato. The rate of red color development will be determined by the curve showing the relationship between the CIE a* value and storage period. Furthermore, the standard curve for predicting the red color development will be developed according to the relationship between CIE a* value and storage period that was approached by the curve fitting method to sigmoid function. The preliminary study showed that the lag time of tomato was a temperature dependent. The higher the temperature, the shorter the lag time and vice versa. This trend was also observed for the rate of red color development. By using these data including the data from the ongoing experiment, standard curves of red color development of mature green tomato during ripening at different storage temperature conditions will be developed. It will enable us to simply predict the ripening stage of tomato harvested at the mature green stage based on the temperature and storage period.

Oral Session | Postharvest/Food Technology and Process Engineering
[4-1600-C] Postharvest/Food Technology and Process Engineering (4)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C (3rd room)

[4-1600-C-04] Quality and Shelf-life Prediction of Fresh-cut 'Phulae' Pineapple by Using Image Analysis and Artificial Neural Networks

*Rattapon Saengrayap[1], Mayura Dongsuea[1] （1. Postharvest Technology and Logistics Program, School of Agro-Industry, Mae Fah Luang University(Thailand)）
Keywords: Browning index, Fractal dimension, Image features, Image processing, Storage

The image analysis technique had been applied for determining quality of fruit as a low cost, fast, and effective technique. In this study, the extracted image features, i.e., color, size, and texture, were then used as a criterion for predicting fresh-cut fruit quality and shelf-life. The aim of this study was to develop the suitable artificial neural network (ANN) model for predicting quality and shelf-life of fresh-cut 'Phulae' pineapple by using the information from image analysis. A green-yellow maturity stage of 'Phulae' pineapple [*Ananas comosus* (L.) *Merr.*] were cut into a cubical shape of $2\times2\times2$ cm^3 and stored at 5 and 10°C for 0, 2, 4, 6, and 8 days. The color (CIELAB values), shrinkage coefficient and firmness of fresh-cut 'Phulae' pineapple were determined every two days. The results showed that a higher storage temperature had a strong influence on the change of color. The *L** values decreased since browning occurred resulted in the increased of browning index (BI). Moreover, the greater storage temperature also influenced the higher change of fruit shrinkage and firmness. The texture of the fresh-cut pineapple became softer and shrinkage was obviously observed as storage time increased. On the other hand, the image analysis technique was also used to assess the quality of fresh-cut 'Phulae' pineapple. The change of RGB color values, fruit dimension, and fractal dimension (FD) value were determined. The results showed that the R (red) value increased as the browning occurred. Moreover, a large variation of browning intensity and its area on the pineapple surface resulted in a larger FD value. The multi-layer feed-forward back propagation ANNs were developed to predict quality and shelf-life of fresh-cut 'Phulae' pineapple. The inputs of the model were storage temperature, fruit dimension, R and FD values, while the outputs were BI, shrinkage coefficient, and shelf-life. The numbers of the hidden node in a hidden layer were varied from two to forty with the increment of two. According to the selection of the best model for predicting the quality and shelf-life, the 18 hidden-node architecture was the most suitable model which provided R^2 of 0.98 and mean square error (MSE) of 0.01, 0.002 and 0.2 day for BI, shrinkage coefficient, and shelf-life, respectively.

Quality and Shelf-life Prediction of Fresh-cut 'Phulae' Pineapple by Using Image Analysis and Artificial Neural Networks

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ABSTRACT

The image analysis technique had been applied for determining quality of fruit as a low cost, fast, and effective technique. In this study, the extracted image features, i.e., color, size, and texture, were then used as a criterion for predicting fresh-cut fruit quality and shelf-life. The aim of this study was to develop the suitable artificial neural network (ANN) model for predicting quality and shelf-life of fresh-cut 'Phulae' pineapple by using the information from image analysis. A green-yellow maturity stage of 'Phulae' pineapple [*Ananas comosus* (L.) *Merr.*] were cut into a cubical shape of $2\times2\times2$ cm^3 and stored at 5 and 10°C for 0, 2, 4, 6, and 8 days. The color (CIELAB values), shrinkage coefficient and firmness of fresh-cut 'Phulae' pineapple were determined every two days. The results showed that a higher storage temperature had a strong influence on the change of color. The *L** values decreased since browning occurred resulted in the increased of browning index (BI). Moreover, the greater storage temperature also influenced the higher change of fruit shrinkage and firmness. The texture of the fresh-cut pineapple became softer and shrinkage was obviously observed as storage time increased. On the other hand, the image analysis technique was also used to assess the quality of fresh-cut 'Phulae' pineapple. The change of RGB color values, fruit dimension, and fractal dimension (FD) value were determined. The results showed that the R (red) value increased as the browning occurred. Moreover, a large variation of browning intensity and its area on the pineapple surface resulted in a larger FD value. The multi-layer feed-forward back propagation ANNs were developed to predict quality and shelf-life of fresh-cut 'Phulae' pineapple. The inputs of the model were storage temperature, fruit dimension, R and FD values, while the outputs were BI, shrinkage coefficient, and shelf-life. The numbers of the hidden node in a hidden layer were varied from two to forty with the increment of two. According to the selection of the best model for predicting the quality and shelf-life, the 18 hidden-node architecture was the most suitable model which provided R^2 of 0.98 and mean square error (MSE) of 0.01, 0.002 and 0.2 day for BI, shrinkage coefficient, and shelf-life, respectively.

Keywords: Browning index, Fractal dimension, Image features, Image processing, Storage

1. INTRODUCTION

'Phulae' pineapple [*Ananas comosus* (L.) *Merr.*] is one of the economical plants in Chiang Rai province. In a recent year, 'Phulae' pineapple has the highly domestic and oversea demand, especially an enormous Chinese market. Fresh-cut produces tend to satisfy the needs of consumer for a freshly convenient prepared food (Sillani & Nassivera, 2015). However, the fresh-cut produces may suffer with a rapid decrease of a quality, especially, their appearance, texture, and flavor (Hurling and Shepherd, 2003; Salvador et al., 2007; Wu and Sun, 2013; Allegra et al., 2015). To ensure the quality attributes of product, the quality evaluation techniques are concerned. Non-destructive evaluation is proposed as an alternative method to assess the quality without any invasive to product. Image analysis has been archived to estimate the external appearances such as size, shape and color of agricultural products (Hussain, Pu, & Sun, 2018; Moreda, Ortiz-Cañavate, García-Ramos, & Ruiz-Altisent, 2009).

In fresh-cut produce, image analysis techniques have been developed for assessing product quality and shelf-life (Pace et al., 2014; Cáez Ramírez et al., 2017; Cavallo et al., 2018). Image features extracted from the digital image were used as a key information for estimating product quality, i.e., color (Gomes et al., 2014; El-Bendary et al., 2015; Mohammadi et al., 2015), size (Moreda et al., 2009; Opara and Pathare, 2014; Ullah et al., 2018; Calixto et al., 2019), shape (Mollazade, Omid, & Arefi, 2012; Yang, Zhang, Zhai, Pang, & Jin, 2019), and texture (Borah, Hines, & Bhuyan, 2007; Pongmalai, Devahastin,

Chiewchan, & Soponronnarit, 2015; R. Quevedo, Pedreschi, Bastias, & Díaz, 2016; Roberto Quevedo, Díaz, Caqueo, Ronceros, & Aguilera, 2009).

Artificial neural network (ANN) model has been extensively achieved as a prediction tool in many studies. This is because of its ability to learn a non-linear complex relationship between input and output. The application of ANN has been found in a various fields, including crops yield prediction (Naroui Rad, Ghalandarzehi, & Koohpaygani, 2017; Naroui Rad, koohkan, Fanaei, & Pahlavan Rad, 2015; Soares, Pasqual, Lacerda, Silva, & Donato, 2013), fresh produce quality estimation (Sanaeifar, Bakhshipour, & de la Guardia, 2016; Zarifneshat et al., 2012), grading and sorting (De Oliveira, Leme, Barbosa, Rodarte, & Alvarenga Pereira, 2016; Mollazade et al., 2012; Wan, Toudeshki, Tan, & Ehsani, 2018), etc. ANN has also been reported to use for shelf-life estimation of fruits and vegetables (Mohi Alden, Omid, Rajabipour, Tajeddin, & Soltani Firouz, 2019; Sanaeifar et al., 2016), however, the information were still limited.

Therefore, in this study, the image analysis was proposed as a tool for extracting important quality information from the digital image. The image features, i.e., color, size, and texture, were used as the inputs for developing the ANN model. The objective of this study was to develop the suitable ANN model for predicting quality and shelf-life of fresh-cut 'Phulae' pineapple by using the information from image analysis.

2. MATERIALS AND METHODS

A green-yellow maturity stage of 'Phulae' pineapple [*Ananas comosus* (L.) *Merr.*] was harvested at Nanglae district, Chiang Rai Province, Thailand. Pineapple fruits were graded with the criteria of 50% yellow peel colored and the uniform size of 250-300g. The fruits then thoroughly washed, peeled, and removed the eye of the fruit. The peeled pineapples cut into a cubical shape of $2 \times 2 \times 2$ cm^3. The cut fruits were packed into the Styrofoam trays covered with plastic wrap and stored at 5 and 10°C prior to determine the qualities at day 0, 2, 4, 6, and 8.

2.1 Quality Determination

2.1.1 Color

Color of 'Phulae' pineapples were evaluated by colorimeter (MiniScan EZ, USA) with settings standard based on illuminant/Observer of D65/10º. The CIELAB values, i.e., lightness (L^*), redness (a^*), and yellowness (b^*) were measured and recorded using Easy Match QC 4.70 software (version 2.2, USA) from three different positions. The change of fresh-cut pineapple color was reported in terms of browning index (BI) that calculated using the following equation:

$$BI = [100(x - 0.31)/0.71] \tag{1}$$

where $x = (a^* + 1.75L^*)/(5.645L^* + a^* - 3.012b^*)$

2.1.2 Shrinkage

Shrinkage of fruit was determined using toluene displacement method (Saengrayap et al., 2014) and reported in terms of shrinkage coefficient. Three pieces of cut pineapple were soaked into toluene that the weight was prior measured. Recording the change in weight of toluene, then calculated volume of product by the following equation:

$$V = m/\rho \tag{2}$$

where V is volume of fresh-cut pineapple (m^3), m is weight of toluene (kg) and ρ is density of toluene (862.27 kg/m^3). Then, the shrinkage coefficient was calculated using the following equation:

$$Shrinkage\ coefficient\ (SC) = 1-(V/V_0) \tag{3}$$

where V is the volume of fresh-cut pineapple (mm^3) and V_0 is the initial volume of fresh-cut pineapple at day 0 (mm^3).

2.1.3 Firmness

Firmness of fresh-cut pineapple was measured as puncture force by a texture analyzer (TA-XT Plus; Stable Micro Systems, UK) using 2 mm cylindrical probe. The speed of 0.5 mm/s and penetration distance of 3 mm was used to measure the samples. The maximum force values (N) were reported from the mean value of three replicates.

2.2 Image Analysis

2.2.1 Image Acquisition

A lightbox with installed LED lamp (Philips, TL-D Deluxe, natural day light 18W) was used for imaging. The images were snapped from top of the box with the distance of 50 cm between sample and camera lens. The DSLR camera (Canon EOS – 450D, Tokyo, Japan) was used for capturing image with a

manual mode, auto focus, lens aperture at f 8.0, with 1/50 shutter speed and ISO 200. The original image files (4,288×2,848 pixels) were saved into JPEG format with the resolution of 300 dpi.

2.2.2 Image Pre-processing

Image analysis was performed by using 'ImageJ' software (version 1.51j8, NIH, Bethesda, MD, USA). The original images were cropped and resized in 400×600 pixels then whole background and noises were removed prior to analyze.

2.2.3 Color

RGB color values were extracted from pineapple images and then converted into CIELAB using color CIE standard equations (León, Mery, Pedreschi, & León, 2006). The performance for the use of image analysis and the total color difference (ΔE) were then calculated. The converted CIELAB values were then compared with the actual values obtained from colorimeter by using the following equation:

$$\Delta E = [(L_c^*-L^*) + (a_c^*-a^*) + (b_c^*-b^*)]^{1/2} \tag{4}$$

where L^*, a^*, and b^* were the actual lightness, redness and yellowness value from colorimeter, and L_c^* a_c^*, and b_c^* were the converted lightness, redness, and yellowness values.

Simple linear regressions were calculated to analyze the relationship by using correlations between converted CIELAB and colorimeter CIELAB. The RGB values were also determined a relationship between among CIELAB value using Pearson's correlation coefficient analysis at significant level ($P < 0.01$).

2.2.4 Dimension

Dimensions of fruit were determined by using 'analyze particle' function. The cropped RGB images were converted into gray scales and binary images. The major and minor dimensions were then determined and reported in the unit of mm.

2.2.5 Fractal Dimension

Fractal dimension (FD) values were determined by converting cropped RGB images into 8-bit gray scale images. The surface plot of the 8-bit gray scale images was performed to see the intensity of browning incidence. The images were then converted into a binary image prior to determine FD values. A box counting method was applied for determining FD values (Roberto Quevedo et al., 2009).
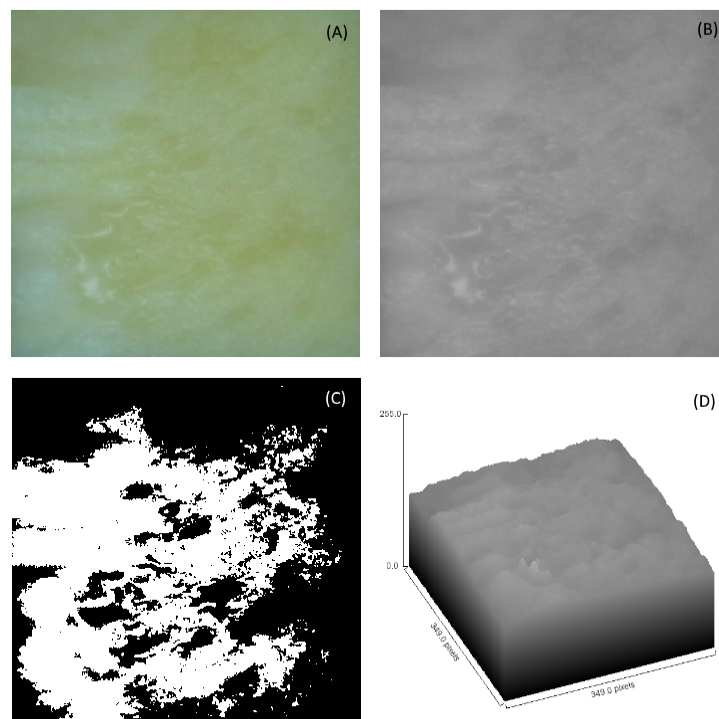
Figure 1. Image analysis procedure for FD determination: (A) — cropped RGB image; (B) — gray scale image; (C) — binary image; and (D) — surface plot.

2.3 ANN Development

ANN modelling was performed for quality and shelf-life prediction by using R (version, 3.5.0, 64 bits) and RStudio (version, 1.1.447) software. The one hidden layer feed forward back propagation architecture ANN was developed. Five different characteristics including storage temperature (T), fruit dimension (D), red (R) and FD values (FD) were used as inputs, while browning index (BI), shrinkage coefficient (SC), and shelf-life (t) was assigned as output. Moreover, the different numbers of hidden node were also tested (2-40 nodes with the increment of 2). The typical diagram of the developed ANN is shown in Figure 2
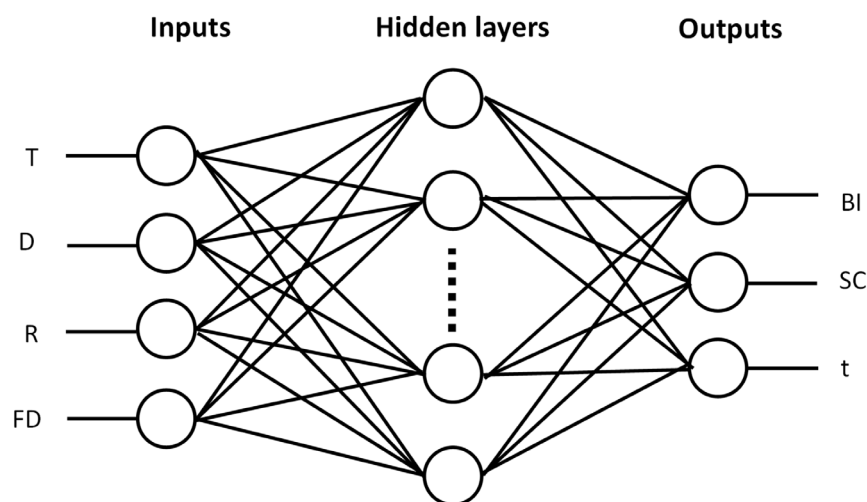
Figure 2. The typical diagram of developed ANN: T — storage temperature; D — fruit dimension; R — red value; FD — FD value; BI — browning index; SC — shrinkage coefficient; and t — shelf-life.

2.3.1 ANN performance

The selection of the best ANN for predicting quality and shelf-life of pineapple was based on their coefficient of determination (R^2) and mean square error (MSE) values. The best model should provide a highest R^2 and lowest MSE.

3. RESULTS AND DISCUSSION

3.1 Color

The significant change of redness (a^*) was found in fresh-cut pineapple stored at 10°C. The increased of a^* value regarding to the occurance of browning reaction during storage. Moreover, the lightness (L^*) of fresh-cut pineapple stored at 10°C was also gradually decreased as browning increased. In terms of lower storage temperature, all color parameters seemed to be constant with no significant change (Figure 3A-C). The results of browning index (BI) over storage time is shown in Figure 3D. The BI of 10°C stored pineapple showed the higher trend compared with those of 5°C.
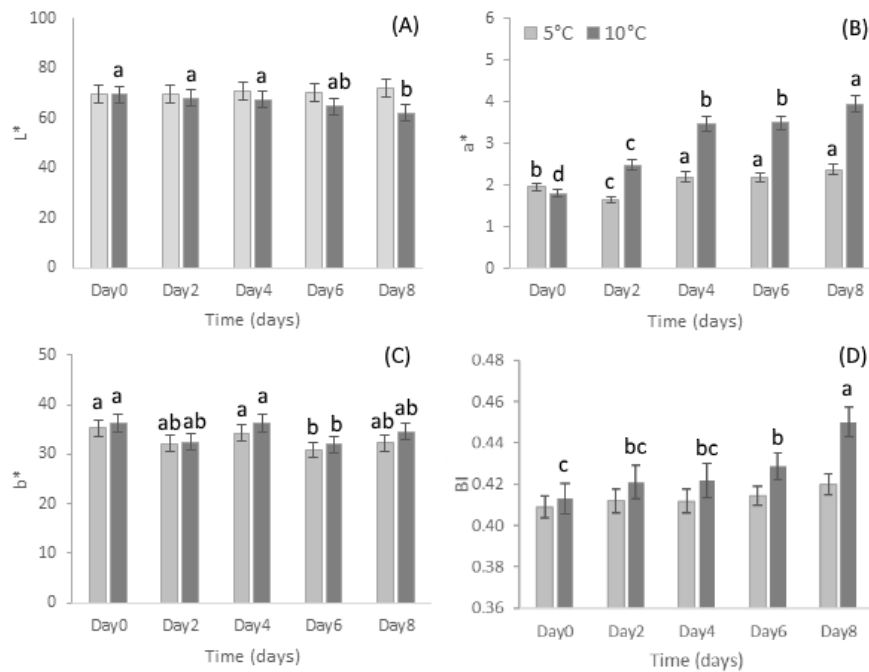
Figure 3. Effect of storage time and temperature on the change of fresh-cut pineapple color during storage: (A) — lightness; (B) — redness; (C) — yellowness; (D) — browning index.

3.2 Shrinkage

The results showed that the shrinkage coefficient of fresh-cut pineapple increased in all storage condition during storage (Figure 4A). At day 6, fresh-cut pineapple stored at 10°C had a significant higher shrinkage value compared with those of 5°C. Since water losses resulted to the change of cell structure. The internal structure collapsed led to the surface wilting and external structure shrinkage. A higher rate of shrinkage in fresh-cut fruit are also reported by a higher rate of water losses (Paniagua, East, Hindmarsh, & Heyes, 2013).

3.3 Firmness

The firmness of fresh-cut pineapple decreased after day 2 of storage in both storage conditions and remained constant (Figure 4B). At 5°C, the firmness seemed to be higher, but did not significant difference. Water losses might result in the change of fruit shrinkage, however, the fibrous surface of pineapple that exposed to the cold air seemed to dry and performed like a hard crust the firmness of fresh-cut pineapple was then remained constant during time of storage (Gallotta, Allegra, Inglese, & Sortino, 2018; Tirkey et al., 2014).
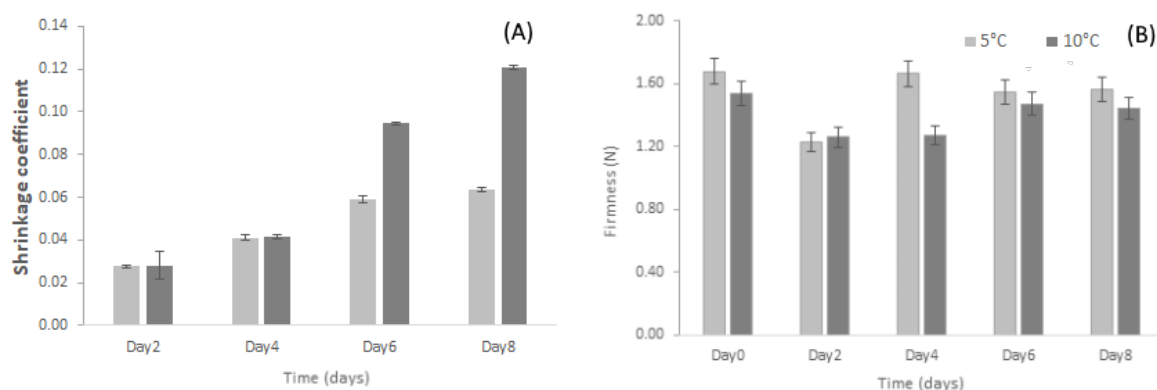
Figure 4. Effect of storage time and temperature on the shrinkage coefficient (A) and (B) firmness of fresh-cut pineapple.

3.4 Color

The results showed that the red (R) values significantly increased with the storage time, while the green (G) and blue (B) values were constant. The increased of browning on fresh-cut pineapple surface resulted in the increasing of R value. The increased of R value during storage was similar to the increased of *a** value (Cáez Ramírez et al., 2017; Nadafzadeh, Abdanan Mehdizadeh, & Soltanikazemi, 2018; Sanaeifar et al., 2016). Storage temperature was also had an influence on the change of R value. A greater increased of R value was found at storage temperature of 10°C. The converted CIELAB values showed a good agreement with those of actual value from colorimeter (Figure 5). The linear regressions showed a good fit between the converted and actual CIELAB values (R^2 >0.93). Moreover, the total color difference (ΔE) of this study was 3.54. The lower ΔE values showed the accuracy of the developed image analysis procedure (León et al., 2006).
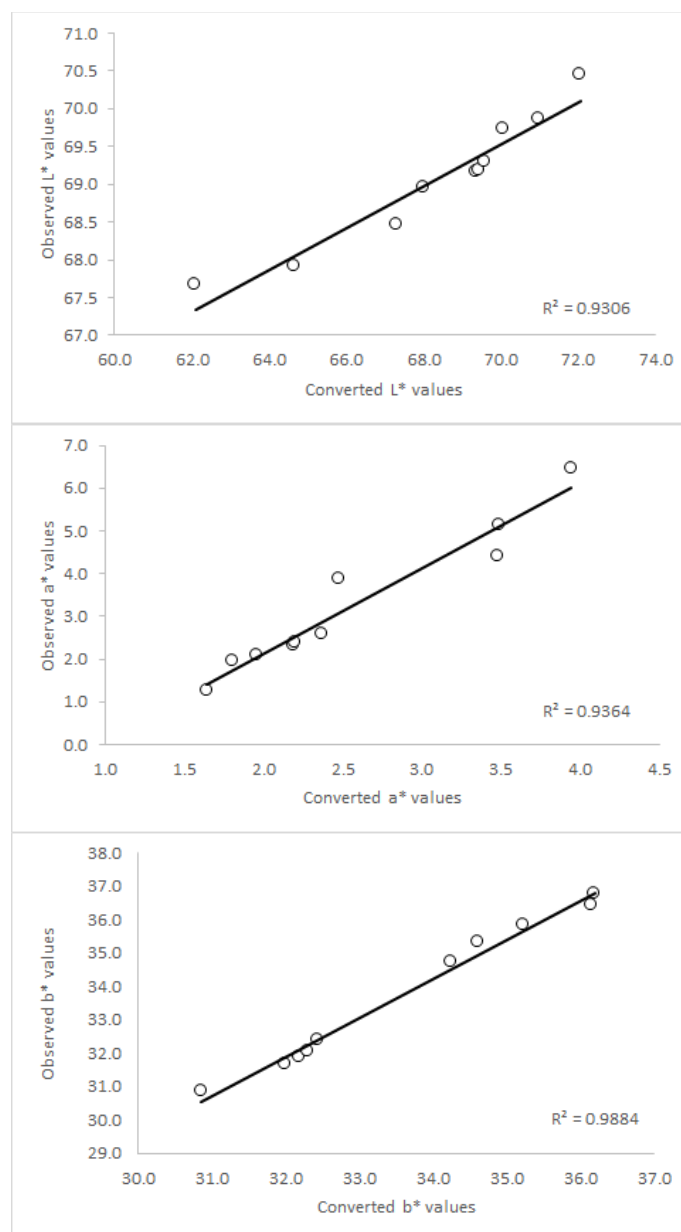
Figure 5. Comparison between converted and actual CIELAB values.

The correlation coefficients between the extracted RGB values, actual CIELAB of fresh-cut pineapple were investigated. Table 1 shows the significant correlation coefficient of RGB values with CIELAB values. According to the results, it indicated that R value was the best parameter for evaluating browning of fresh-cut pineapple.

Table 1. Peason's correlation coefficients between RGB values and CIELAB values.

Parameters	R	G	B	L^*	a^*	b^*
R	1.000					
G	0.577*	1.000				
B	0.328	0.468	1.000			
L^*	-0.921**	0.744**	0.696*	1.000		
a^*	0.934**	0.788**	0.639*	-0.800**	1.000	
b^*	-0.805**	0.738**	0.703**	0.533*	0.614*	1.000

**, * Denotes significant correlation coefficients at (P<0.01) and (P<0.05) by Peason's correlation test.

3.5 Dimension

The dimension reduction during storage was significant detected by image analysis. Table 2 shows the results of area and dimension reduction of fresh-cut pineapple during storage at 5 and 10°C. Both surface area and dimension were significantly reduced during storage. This may occur because of loss of water. Higher storage temperature caused fast shrinkage of fruit (Mahajan, Oliveira, & Macedo, 2008; Murmu & Mishra, 2016). The dimension reduction trend that observed from image analysis had a good agreement with those of shrinkage coefficient value.

Table 2. Effect of storage time and temperature on the change of fresh-cut pineapple's surface area and dimension.

Temperature	Time	Dimension reduction	Surface area reduction
°C	Day(s)	%	%
5	0	0.00 ± 0.00^e	0.00 ± 0.00^e
	2	1.54 ± 0.17^d	3.06 ± 0.37^d
	4	2.61 ± 0.20^c	5.15 ± 0.21^c
	6	3.79 ± 0.29^b	7.44 ± 0.36^b
	8	4.15 ± 0.14^a	8.14 ± 0.22^a
10	0	0.00 ± 0.00^d	0.00 ± 0.00^e
	2	2.38 ± 0.38^c	4.70 ± 0.66^d
	4	3.42 ± 0.23^b	6.72 ± 0.54^c
	6	4.32 ± 0.23^a	8.45 ± 0.24^b
	8	4.78 ± 0.28^a	9.33 ± 0.48^a

3.6 Fractal dimension

The results showed that fractal dimension values (FD) decreased during storage at 5°C. On the other hand, higher storage temperature of 10°C, the FD values found to be significantly increased (Table 3).

Table 3. Effect of storage time and temperature on the change of fractal dimension (FD) of fresh-cut pineapple.

Time	Temperature	
Day(s)	5°C	10°C
0	1.73 ± 0.02^a	1.67 ± 0.01^c
2	1.70 ± 0.07^a	1.67 ± 0.03^c
4	1.69 ± 0.02^{ab}	1.75 ± 0.01^b
6	1.68 ± 0.02^{ab}	1.77 ± 0.03^b
8	1.67 ± 0.04^b	1.83 ± 0.02^a

The decreasing of FD value was reported in fresh-cut fruit during storage since the uniform color distribution on fruit surface. Figure 7A-7E shows the RGB image of fresh-cut pineapple, uniformly change of surface color resulted in constant FD value after day 2 (1.67-1.70). The heterogeneous distribution of the color surface resulted in the increasing of FD value (R. Quevedo et al., 2016; Roberto Quevedo et al., 2009). The ununiform browning reaction on pineapple surface at 10°C might influenced the increasing of FD value (Figure 7K-7O).
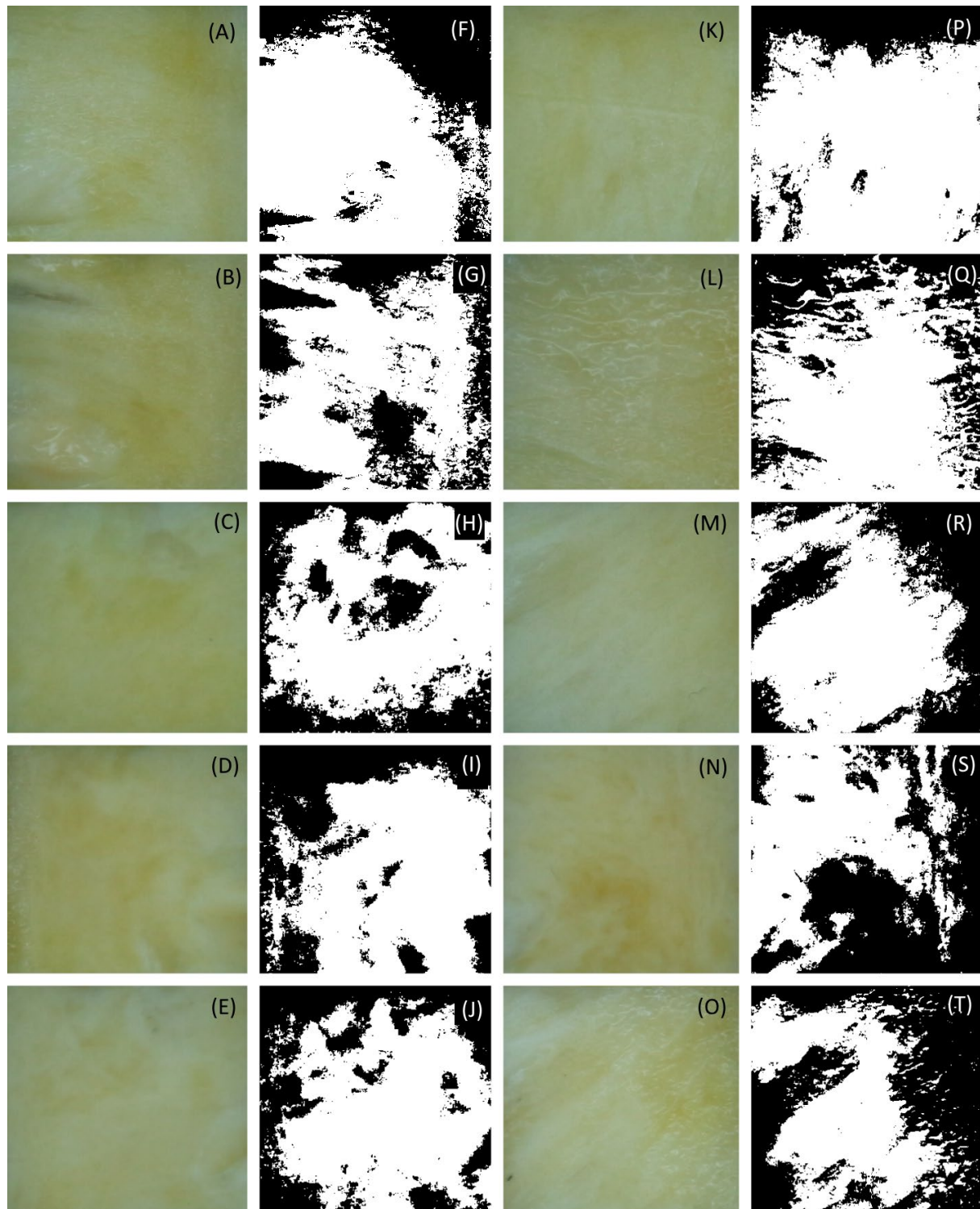
Figure 7. RGB images of fresh-cut pineapple stored at 5°C (A – day 0; B – day 2; C – day 4; D – day 6; and E – day 8) and 10°C (K – day 0; L – day 2; M – day 4; N – day 6; and O – day 8) and their corresponding binary image (5°C — F-J; 10°C — P-T) during storage.

3.7 ANN for qualities and shelf-life prediction

The performance of developed ANN shows in Table 4. The results showed that the 18-hidden node architecture provided the best prediction result with R^2 of 0.98 and MSE of 0.01, 0.002 and 0.2 day for browning index (BI), shrinkage coefficient (SC), and shelf-life (t), respectively. The results also showed that the increasing in number of the hidden node resulted in decreasing of prediction accuracy. A larger

number of hidden nodes increased the complexity of model architecture which lead to the increasing in prediction error (Mohi Alden et al., 2019; Shabani, Ghaffary, Sepaskhah, & Kamgar-Haghighi, 2017; Suárez Salazar, Melgarejo, Durán Bautista, Di Rienzo, & Casanoves, 2018). The accuracy of the developed model might decrease when the number of hidden node exceeded the optimum number.

Table 4. Prediction performance of developed ANN for predicting fresh-cut pineapple qualities and shelf-life.

Number of hidden node	R^2	Mean square error (MSE)		
		BI	SC	t (days)
2	0.88	0.05	0.010	1.0
4	0.86	0.05	0.010	1.0
6	0.89	0.05	0.009	0.9
8	0.91	0.03	0.006	0.6
10	0.93	0.02	0.004	0.4
12	0.94	0.02	0.004	0.4
14	0.92	0.02	0.003	0.3
16	0.95	0.02	0.003	0.3
18	0.98	0.01	0.002	0.2
20	0.95	0.02	0.003	0.3
22	0.96	0.02	0.003	0.3
24	0.93	0.02	0.004	0.4
26	0.93	0.02	0.004	0.4
28	0.92	0.02	0.003	0.3
30	0.92	0.02	0.003	0.3
32	0.91	0.04	0.008	0.8
34	0.88	0.05	0.009	0.9
36	0.88	0.05	0.010	1.0
38	0.86	0.06	0.011	1.1
40	0.88	0.06	0.011	1.1

4. CONCLUSION

The change of physical properties of fresh-cut 'Phulae' pineapple during cold storage was observed using both conventional and developed image analysis techniques. Higher storage temperature affected a higher change of all physical properties including color, shrinkage, firmness. The developed image analysis technique showed the possibility to determine the quality of fresh-cut as expected. The R and FD values were able to explain the occurrence of browning reaction occurred during storage. The dimension reduction obtained from image analysis could explain the change of product shrinkage with good agreement. The developed 18-hidden node ANN model with the selected inputs form image analysis including storage temperature, fruit dimension, R and FD values was able to estimate shelf-life, shrinkage and browning index of fresh-cut pineapple with a high accuracy ($R^2 > 0.98$).

ACKNOWLEDGMENT

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Oral Session | Postharvest/Food Technology and Process Engineering
[4-1600-C] Postharvest/Food Technology and Process Engineering (4)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C (3rd room)

[4-1600-C-05] Stationary Machine Vision Based Real-Time Estimation of Japanese Black Cattle Serum Vitamin A using Eye Fundus Color

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Keywords: Japanese Black Cattle, Beef, Vitamin A, Fundus Color, Machine Vision

Japanese black cattle are well known all over the world for their highly tender and deep flavored beef. In Japan, Beef Marbling Standards (BMS) has predominantly become the most influential factor for deciding the beef quality. Due to the negative correlation between serum vitamin A (Vit. A) level and BMS, higher BMS has been achieved by gradually reducing the amount of serum Vit. A in the cattle during fattening. This activity can be dangerous therefore needs to be precisely monitored. The conventional way of monitoring Vit. A level is by blood sampling and it`s very costly, time-consuming, and stressful to the cattle. As a result, our laboratory members have tirelessly conducted a number of researches to develop the best alternative. These previous researches have achieved Vit. A level estimation from the eye surface features like pupil color, pupil reflection, and pupil light reflex. In this research, we investigate the eye fundus color in addition to the pupil color, pupil reflection, and pupil light reflex in a stationary machine vision system to estimate serum Vit. A level in Japanese Black Cattle.

Stationary Machine Vision Based Real Time Estimation of Japanese Black Cattle Serum Vitamin A using Eye Fundus Color

oSamuel Ouma[1], Naoshi Kondo[1], Tateshi Fujiura[1], Yuichi Ogawa[1], Tetsuhito Suzuki[1], Katsuya Takenouchi[1], Hidetsugu Yoshioka[1], Moriyuki Fukushima[2], Takahiko Ohmae[3]

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[Introduction]

Japanese black cattle are well known for their highly tender and deep flavored beef. To meet consumers demand, the Japanese farmers consistently add value by producing high quality beef. The degree of marbling (BMS) has predominantly become the most influential factor for deciding the quality. This is because of the slight increase in flavor, juiciness, and tenderness associated with.

Due to the negative correlation between serum vitamin A (vit. A) level and BMS, higher BMS have been achieved by gradually reducing the amount of serum vit. A in the cattle during fattening [2]. However, vit. A level lower than 30IU/dL is dangerous to cattle health [1]. Hence, the need to monitor vit. A level during fattening.

The convectional way of monitoring vit. A level is by blood sampling which is very costly, invasive, time consuming, and stressful to the cattle. As a result, our laboratory members have over the years conducted a number of researches to develop the best alternative. These researches have achieved vit. A level estimation from the eye surface features which includes pupil color, pupil reflection, and pupil light reflex.

However, these methods rely to a large degree average reflection from the pupil when an incident light interacts with the inside of the eye. Because of diffused reflection inside the eye, the image of the pupil area is usually not clear.

To improve the image quality, a fundus image (inner lining of the eye) and additional color feature of the fundus were acquired for vit. A level estimation. Another advantage of adding fundus color changes are that they are a consistent sign compared to other ocular changes[2]. These changes reflect the general systemic level of vit. A.

[Experimental Method

Four Japanese Black cattle raised in Tajima Agricultural High School; Hyogo Prefecture from May 2017 until June 2019 were used. The vit. A level measured by blood assay in these cattle ranged from 14 to 98 IU/dL. An automatic image acquisition system was developed and installed at the cattle drinking place. When system detects the cattle drinking, cattle's eye is illuminated by white LED and the tapetum image is captured.

The fundus area was automatically extracted from the original image using C++ program and OpenCV library (Fig. 1). Fundus color feature was analyzed and the relationship between vit. A with r, g, and b component ratios were investigated.
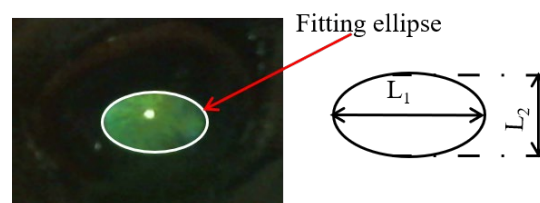
Fig. 1 Image of the fundus and the fitting ellipse

The Red (r), Blue (b), and Green (g) component ratios were calculated by the formulas:

$r=R/(R+G+B)$, $b=B/(R+G+B)$, $g=G/(R+G+B)$

[Results and Discussion]
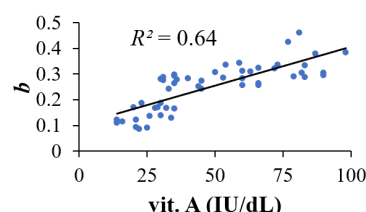
The b component (Fig. 2) values had a clear

Fig. 2 Relationship of and b ratios with vit. A level

positive correlation ($R^2 = 0.64$) with vit. A level changes. While on the other hand, the r component values showed a negative correlation ($R^2 = 0.58$) with vit. A. Cattle eye fundus is typically tapetal. Under normal circumstances of vit. A, the tapetum in cattle appears greenish-blue in color[3]. Under vit. A deficiency, the tapetum became bleached.

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5:15 PM - 5:30 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C)

[4-1600-C-06] Segmentation of common scab lesion on intact potatoes using single near-infrared image

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Keywords: image processing, common scab, external defects , near infrared, segmentation

Segmentation and identification of potato common scab lesion on a single near infrared image is the objective of this research. A more simple machine vision system is desired for sorting system application. Thus, a minimum number of the image is preferable compared to multispectral images for speediness if it can give good results. In this research, we proposed an imaging system using an InGaAs camera and a bandpass filter at 1600 nm which found to provide a good contrast of common scab lesion to normal potato skin. An image correction method was employed to deal with uneven light distribution due to the various shape of potatoes. Image segmentation has been successfully carried out with a Dice Sorensen coefficient of 0.72. Results also show that external defects, such as common scab and some mechanical damage types, and soil deposits appear brighter in the near infrared region, against the normal skin background. Thus, further image analysis of area shape and textural features has been provided to discriminate common scab and other classes. The results could be used as consideration for a real sorting application.

5:30 PM - 5:45 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C)

[4-1600-C-07] Myanmar Mango Maturity Prediction Based on Skin Color Using Machine Vision System

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Keywords: Maturity prediction, Machine vision

Mango fruits mostly in Myanmar, are harvested without maturity grading, which will cause later problems in processing and transportation. Mangoes of different maturity level show difference in skin characteristics like color. As machine vision system can acquire visual information of an object and give feedback in a short time, it is likely to be an effective method to predict mangoes' maturity based on their skin characteristics. In this study, to better evaluate mangoes' skin characteristics, 3 color models (RGB, HSV, CIELAB) were compared and discussed. Sein ta Lone mangoes' skin characteristics during maturation were recorded as color images and fluorescence images using USB camera with white and UV LED illumination. Changes of color figures in 3 color models of 2 types of images during maturation were analyzed using Mat lab and compared. As a result, a* value from the CIELAB color model is observed an obvious change during mango maturation in color images compared to others.

Oral Session | Postharvest/Food Technology and Process Engineering
[4-1600-C] Postharvest/Food Technology and Process Engineering (4)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C (3rd room)

[4-1600-C-08] Measurement of Chicken Eggshell Optical Properties Using Terahertz Spectroscopy

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Keywords: Eggshell, Optical properties, Refractive index, Dielectric constant, Extinction co-efficient, Terahertz spectroscopy

Terahertz (THz) is a relatively new and under explored part of the electromagnetic spectrum that promises to be an extremely useful tool for research in the agricultural sciences. One such under explored area is how it interacts with biological objects like avian eggs. In this respect, chicken eggs are considered an ideal representative for avian studies. Eggshell plays various important roles such as protecting the internal contents and embryo of these eggs from ultraviolet (UV) rays, predators and contaminants. In addition, it provides major minerals for embryo development and facilitates embryonic respiration (i.e. gas exchange) during incubation. Electromagnetic waves below the THz range are limited in their ability to probe eggshell properties due to high absorbance and scattering effects in the UV, Visible and even NIR regions. Moreover, eggshell color pigment, called protoporphyrin, masks measurements in the Visible region. Therefore, we aim to measure the optical properties of the chicken eggshell using THz waves to obtain foundational data for researchers working on avian and reptile eggs. Moreover, this dataset could help to inform scientists regarding eggshell factors relevant to their research protocols. Optical properties of chicken (layer) eggshell, such as refractive index, dielectric constant and extinction co-efficient, were measured using Terahertz Time domain Spectroscopy (0.2 to 3.0 Terahertz). THz transmission and THz reflection were measured for broken eggshell and intact eggshell respectively. The refractive index of the eggshell increased slightly with increasing THz frequency; varying from 2.7 to 3.3. Whereas the extinction co-efficient sharply increased from 0.2 to 2.0 THz. The dielectric constant of the eggshell increased slightly from 7.0 to 10.2 in the 0.2 to 2.0 THz range. This research has shown that when probing the optical properties of crystalline structures with very low water content, such as eggshells, THz waves are an appropriate tool. The foundational properties documented in this research can be used in various applied research fields (e.g. applied optics, ecology, ornithology, evolutionary biology) in the future.

Measurement of Chicken Eggshell Optical Properties Using Terahertz Spectroscopy

Alin Khaliduzzaman[1,3*], Keiji Konagaya[1], Tetsuhito Suzuki[1], Ayuko Kashimori[2], Naoshi Kondo[1], Yuichi Ogawa[1]

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Terahertz (THz) is a relatively new and under explored part of the electromagnetic spectrum that promises to be an extremely useful tool for research in the agricultural sciences. One such under explored area is how it interacts with biological objects like avian eggs. In this respect, chicken eggs are considered an ideal representative for avian studies. Eggshell plays various important roles such as protecting the internal contents and embryo of these eggs from ultraviolet (UV) rays, predators and contaminants. In addition, it provides major minerals for embryo development and facilitates embryonic respiration (i.e. gas exchange) during incubation. Electromagnetic waves below the THz range are limited in their ability to probe eggshell properties due to high absorbance and scattering effects in the UV, Visible and even NIR regions. Moreover, eggshell color pigment, called protoporphyrin, masks measurements in the Visible region. Therefore, we aim to measure the optical properties of the chicken eggshell using THz waves to obtain foundational data for researchers working on avian and reptile eggs. Moreover, this dataset could help to inform scientists regarding eggshell factors relevant to their research protocols. Optical properties of chicken (layer) eggshell, such as refractive index, dielectric constant and extinction co-efficient, were measured using Terahertz Time domain Spectroscopy (0.2 to 3.0 Terahertz). THz transmission and THz reflection were measured for broken eggshell and intact eggshell respectively. The refractive index of the eggshell increased slightly with increasing THz frequency; varying from 2.7 to 3.3. Whereas the extinction co-efficient sharply increased from 0.2 to 2.0 THz. The dielectric constant of the eggshell increased slightly from 7.0 to 10.2 in the 0.2 to 2.0 THz range. This research has shown that when probing the optical properties of crystalline structures with very low water content, such as eggshells, THz waves are an appropriate tool. The foundational properties documented in this research can be used in various applied research fields (e.g. applied optics, ecology, ornithology, evolutionary biology) in the future.

Keywords: Eggshell, Optical properties, Refractive index, Dielectric constant, Extinction co-efficient, Terahertz Spectroscopy

6:00 PM - 6:15 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room C)

[4-1600-C-09] Application of LCA (Life Cycle Assessment) Methodology in Bioethanol Production from Sugar Industry Wastewater (Molasses) – A Case Study in West Java Province, Indonesia

*Agusta Samodra Putra[1,2], Ryozo Noguchi[3], Tofael Ahamed[3]　(1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Research Center for Chemistry, Indonesian Institute of Sciences(Indonesia), 3. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan)）
Keywords: molasses, LCA, sugar industry, bioethanol

Sugar industry plays an important role in Indonesia. Sugar industry wastes consist of biomass waste (sugarcane bagasse) and liquid waste (molasses). To increase the economic and environmental performance in the sugar industry, utilization of molasses for biorefinery product such as bioethanol is the appropriate solution. In this study, the environmental performance of integrated sugar and bioethanol industry in West Java Province was investigated. Life Cycle Inventory of sugar industry in Subang and bioethanol industry in Palimanan, West Java Province, Indonesia was investigated. This LCA study follows a *gate-to-gate* system boundary from the sugar industry to bioethanol production. SimaPro v8.0.5 software was used for LCA calculation with Chain Management by Life Cycle Assessment (CML) as an environmental impact assessment method. Acidification potential (AP), global warming potential (GWP100), eutrophication potential (EP), and human toxicity potential (HTP) were quantified with 1 kg of bioethanol product as a functional unit. Based on LCA approach, environmental impacts for producing 1 kg of bioethanol from molasses are 0.0030 kg SO_2 eq of AP, 0.1929 kg CO_2 eq of GWP100, 0.0004 kg PO_4 eq of EP, and 0.1494 kg 1,4-DB eq of HTP. Utilization of chemicals in the fermentation process gave a significant environmental impact. Sugarcane bagasse waste in this industry was used for heat and power generation that enough to fulfill process energy requirement. Environmental performance improvement can be proposed by using the LCA approach. In this industry, chemical usage in the fermentation process is the main environmental impact contributor.

Oral Session | Others (including the category of JSAM and SASJ)

[4-1600-D] Other Categories (1)

Chair:Satoshi Yamamoto(Akita Prefectural University), Kikuhito Kawasue(University of Miyazaki)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D (4th room)

[4-1600-D-01] **Applicability Of Japanese Standard About The Powered Exoskeleton To Agriculture**

*Masahiro Tanaka[1], Satoru Umeno[1], Yutaka Kikuchi[1]（1. National Agriculture and Food Research Organization(Japan)）
4:00 PM - 4:15 PM

[4-1600-D-02] **Research on an Intelligent Robot Eye-hand System for Harvesting Pumpkin in the Outdoor Condition**

*Liangliang Yang[1], Qian Wang[2], Yohei Hosino[1], Hiroki Ishikuro[1], Ying Cao[1]（1. Kitami Institute of Technology(Japan), 2. Ning Xia University(China)）
4:15 PM - 4:30 PM

[4-1600-D-03] **Handy Type Pig Weight Estimation System Based on Random Forest Algorithm**

*Hsu Lai Wai[1], Kikuhito Kawasue[1], Khin Dagon Win[1], Kumiko Yoshida[2]（1. University of Miyazaki(Japan), 2. KOYO Plant Service(Japan)）
4:30 PM - 4:45 PM

[4-1600-D-04] **Plant Disease Identification using Explainable Features with Deep Convolutional Neural Network**

*Harshana Habaragamuwa[1], Yu Oishi[1], Katu Takeya[1], Kenichi Tanaka[1]（1. National Agriculture and Food Research Organization(Japan)）
4:45 PM - 5:00 PM

[4-1600-D-05] **Sensitivity and Dynamic Analysis of Microalgae Fuel Production System Using LCA**

*Riaru ISHIZAKI[1], Ryozo Noguchi[2], Agusta Samodra Putra[1], Tofael Ahamed[2], Makoto M. Watanabe[3]（1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan), 3. Algae Biomass and Energy System R&D Center, University of Tsukuba(Japan)）
5:00 PM - 5:15 PM

[4-1600-D-06] **An Aerial Weed Detection System for Green Onion Crops Using the You-Only-Look-Once (YOLO) Deep Learning Algorithm**

Addie Ira Borja Parico[1], *Tofael Ahamed[2]（1. College of Agrobiological Resource Sciences, School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan)）
5:15 PM - 5:30 PM

[4-1600-D-07] **A Deep Learning and MSM Machine Learning System for Recognition of Weed Infestation in Cabbage Field Using Unmanned Aerial Vehicle**

*Tofael Ahamed[1], Yan Zhang[1], Linhuan Zhang[1], Ryozo Noguchi[1]（1. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan)）
5:30 PM - 5:45 PM

[4-1600-D-08] **Mallard Navigation Using Unmanned Ground Vehicles, Imprinting, and Feeding**

*Hirokazu Madokoro[1], Satoshi Yamamoto[1], Hanwool Woo[1], Kazuhito Sato[1]　(1. Akita Prefectural University(Japan))

5:45 PM - 6:00 PM

[4-1600-D-09] **Onion Bulb Counting in a Large-scale Field Using a Drone with RTK-GNSS**

*Satoshi Yamamoto[1], Hirokazu Madokoro[1], Yo Nishimura[1], Yukio Yaji[1]　(1. Akita Prefectural University(Japan))

6:00 PM - 6:15 PM

Oral Session | Others (including the category of JSAM and SASJ)
[4-1600-D] Other Categories (1)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D (4th room)

[4-1600-D-01] Applicability Of Japanese Standard About The Powered Exoskeleton To Agriculture

*Masahiro Tanaka[1], Satoru Umeno[1], Yutaka Kikuchi[1] (1. National Agriculture and Food Research Organization(Japan))
Keywords: Powered Exoskeleton, Labor-Saving technology, Assistive technology, ISO 13482, JIS B8456-1

In japan, various types of powered exoskeletons have been developed for long ago such as military, construction, transportation, manufacturing and agriculture and now some of them are in the market. They are expected to reduce a physical load on workers resulting from picking up heavy object or continuous working in a half-sitting posture for long time and free workers from these heavy labors because aging labor population has become a serious problem and elderly people and women have need to perform the heavy labors in Japan. For this reason, the robots coexisting with human that provide services to people and used in same space with human have been developed so far and now it is called service robot, which performs useful task for human and equipment excluding industrial automation applications. Furthermore, Japan proposed safety requirements for personal care robots that is a kind of service robots including powered exoskeleton to ISO and it was published as ISO 13482 on 2014. In this way, Japan is not only paying attention to developing but also establishment of standards about service robots. Especially regarding a powered exoskeleton for lumbar support, a Japanese national standard that specified safety and performance requirements was published as JIS B8456-1 developed from ISO 13482 on 2017. However, it needs to consider details in each field to widely spread through the market, because JIS B8456-1 was cross-cutting standard that summarized the minimum and common requirements in various fields. Therefore, we examined the applicability of this standard to agriculture and there were some problems as a result. Regarding safety requirements, it was considered necessary to identify particular risk factors of using a powered exoskeleton that have the potential to cause harm when farmer would use it in their farm work. Therefore, the authors summarized the risk assessment sheet about using a powered exoskeleton in farm work, so that estimated that unstable surfaces of ground might cause user falling down in particular. Regarding performance requirements, the test methods that measuring assistive torque a powered exoskeleton have were not suitable for agricultural use because it was only for static force however not for dynamic force. Accordingly, the authors developed a measuring instrument and test method for measuring the dynamic assistive torque in consideration of agricultural use. Furthermore, they examined the plasticity using a powered exoskeleton for farm work made in Japan. As a result, the dynamic assistive torque properties were revealed with high reproducibility and standard deviation of maximum and average assistive torque were lower than 1 N・m. the authors expect that risk assessment sheet and measuring instrument they developed can improve applicability of Japanese standard about the powered exoskeleton to agriculture.

Applicability Of Japanese Standard About The Powered Exoskeleton To Agriculture

Masahiro Tanaka*, Yutaka Kikuchi, Satoru Umeno

Institute of Agricultural Machinery, National Agriculture and Food Research Organization, Japan

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ABSTRACT

In Japan, various types of powered exoskeletons have been developed for long ago such as military, construction, transportation, manufacturing and agriculture and now some of them are in the market. They are expected to reduce a physical load on workers resulting from picking up heavy object or continuous working in a half-sitting posture for long time and free workers from these heavy labors because aging labor population has become a serious problem and elderly people and women have need to perform the heavy labors in Japan. For this reason, the robots coexisting with human that provide services to people and used in same space with human have been developed so far and now it is called service robot, which performs useful task for human and equipment excluding industrial automation applications. Furthermore, Japan proposed safety requirements for personal care robots that is a kind of service robots including powered exoskeleton to ISO and it was published as ISO 13482 on 2014. In this way, Japan is not only paying attention to developing but also establishment of standards about service robots. Especially regarding a powered exoskeleton for lumbar support, a Japanese national standard that specified safety and performance requirements was published as JIS B8456-1 developed from ISO 13482 on 2017. However, it needs to consider details in each field to widely spread through the market, because JIS B8456-1 was cross-cutting standard that summarized the minimum and common requirements in various fields. Therefore, we examined the applicability of this standard to agriculture and there were some problems as a result. Regarding safety requirements, it was considered necessary to identify particular risk factors of using a powered exoskeleton that have the potential to cause harm when farmer would use it in their farm work. Therefore, the authors summarized the risk assessment sheet about using a powered exoskeleton in farm work, so that estimated that unstable surfaces of ground might cause user falling down in particular. Regarding performance requirements, the test methods that measuring assistive torque a powered exoskeleton have were not suitable for agricultural use because it was only for static force however not for dynamic force. Accordingly, the authors developed a measuring instrument and test method for measuring the dynamic assistive torque in consideration of agricultural use. Furthermore, they examined the plasticity using a powered exoskeleton for farm work made in Japan. As a result, the dynamic assistive torque properties were revealed with high reproducibility and standard deviation of maximum and average assistive torque were lower than 1 N・m. the authors expect that risk assessment sheet and measuring instrument they developed can improve applicability of Japanese standard about the powered exoskeleton to agriculture.

Keywords: Powered Exoskeleton, Labor-Saving technology, Assistive technology, ISO 13482, JIS B8456-1

1. INTRODUCTION

Japan is one of the robotic powers in the world and the value of shipments of robotics was approximately 900 billion in 2018(Japan Robot Association, 2019). Cabinet Office (2007) estimated that domestic market size of robotics would be 9.7 trillion by 2035 considering future spread of robotics into new fields such as service industry and growth of fields such as manufacturing industry that currently forms the market. Especially, tertiary sector of industry including service industry has been expected to be able to apply robotics in the near future and development of robots for these industries have been promoted.

These robots are called "service robot" and mean robot that performs useful tasks for people or device excluding the use of industrial automation (ISO 13482, 2014). Safety of service robot has been a

problem while expected as major driving force for jump in robot industry in Japan. Sugimoto (2005) mentioned that it was essential to ensure safety of service robot in world-class way to foster robot industry soundly because it might be a sensational event that other machines would not even if an accident rarely occurred. From such a thought, Japan has been considering standardization of safety for service robot since many years ago.

According to Yamada et al. (2007, 2009, 2013), the flow of standardization for service robot is as follows. First of all, discussion about safety for service robot at ISO started in 2002. Hungary proposed standardization activities as robotics application-safety for human rehabilitation within a framework of industrial manipulator at this time, but it was rejected as result of voting by reason that robot for rehabilitation could not be regarded as an application of industrial manipulator. Subsequently, AG (Advisory Group) and 16 WG for mobile servant robot was established in 2005 and the investigation and discussions about NWI (new work item) that should have been targeted as standardization of mobile servant robot been conducted for two years. As a result, PT (Project Team) was established because safety was essential as industries waiting for market expansion and targeted as international standardization that required urgent action. Formulation of standard had been advanced after establishment of WG7 at general meeting of TC 184/SC 2 in 2009 and ISO 13482 was published in 2014 after adopting in FDIS voting.

ISO 13482 regulated three personal care robots out of service robot that performed actions directly to improve the quality of life excluding personal care robot moving at a speed over 20km, underwater robot and flight robot, Industrial robot, robot as medical equipment, robot as military or public power. One of them was physical assistant robot performing assistant or reinforcement of personal physical ability. Requirements for intrinsic safety design, protective measure and information for use were regulated in this. Afterwards, Japanese Industrial Standard for safety requirements of these personal care robots was published as JIS B 8446 and that for safety and performance requirements of powered exoskeleton for lumbar support was published as JIS B 8456-1 in Japan.

JIS B 8456-1 was advanced standard for powered exoskeleton, however, on the other hand, minimum common standard with an eye to the future that new one would appear. Therefore, each industry or fields requires to consider an applicability of this standard to their territory. In this paper, the author considered the applicability of this standard to agriculture and what was necessary for that.

2. MATERIALS AND METHODS

JIS B 8456-1 was generally consist of performance test method and safety requirements for powered exoskeleton. Thus, we considered each applicability to agriculture in the following way.

2.1 Performance test method

JIS B 8456-1 regulated two performance test methods. The one was maximum assistive torque measurement method, the other was disc compression forces and lumbar spine torque measurement method using robot imitating human shape. We actually measured maximum assistive torque of one powered exoskeleton made in Japan following procedure regulated in JIS B 8456-1 that described static torque measurement method, shown Fig1. In summary, the measurer had force gauge and then pull a thigh segment of powered exoskeleton fastened to pedestal while that is working, like tug of war. Although this method was not bad, we confirmed the necessity of dynamic torque measurement to apply this standard to agriculture because body movement farm worker's doing is very fast. Actually, dynamic torque measurement method was considered in the drafting committee of this standard but not regulated because of difficulty and lack of repeatability in testing.

Therefore, we developed a simple and reliable measuring instrument and test method for measuring a dynamic assistive torque generated by the powered exoskeleton and tested functionality of them. Measuring instrument that we developed, shown Fig.2, composed to the fixed pedestal for fastening a powered exoskeleton, an electric slider and the wire with a load cell that connects the powered exoskeleton with the electric slider through a pulley. The powered exoskeleton is fastened to the pedestal shaped like a chair, which was built with a frame and fixed on the floor, by load binding belt. Metal plate substituted for a thigh segment of the powered exoskeleton is bolted to the wire connected to the electric slider, which can move up and down within the range of 500mm stroke, through the pulley. Besides, a load cell attached to the wire provides a pulling force on the wire pulled by metal plate equipped with the powered exoskeleton when it is in operation and metal plate is rotating according to the movement of the electric slider. we examined the repeatability using this instrument.
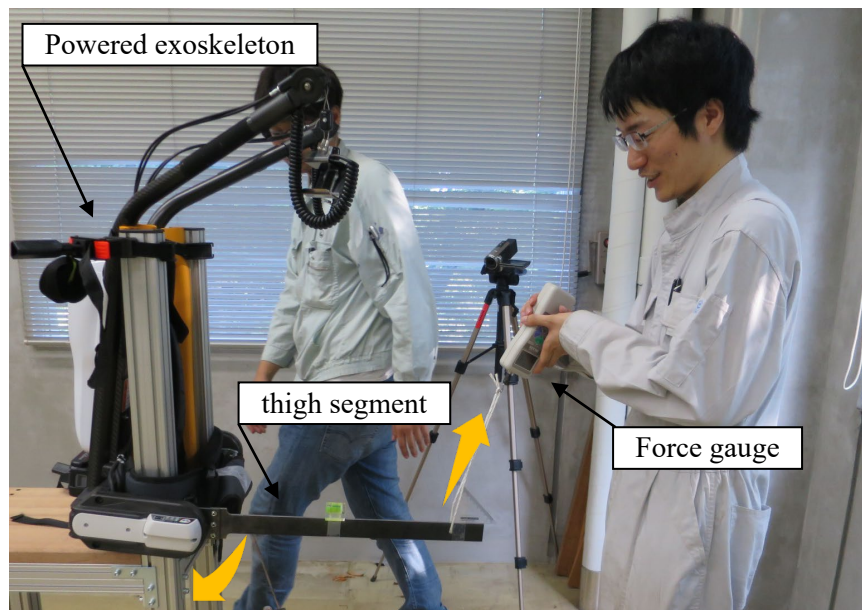
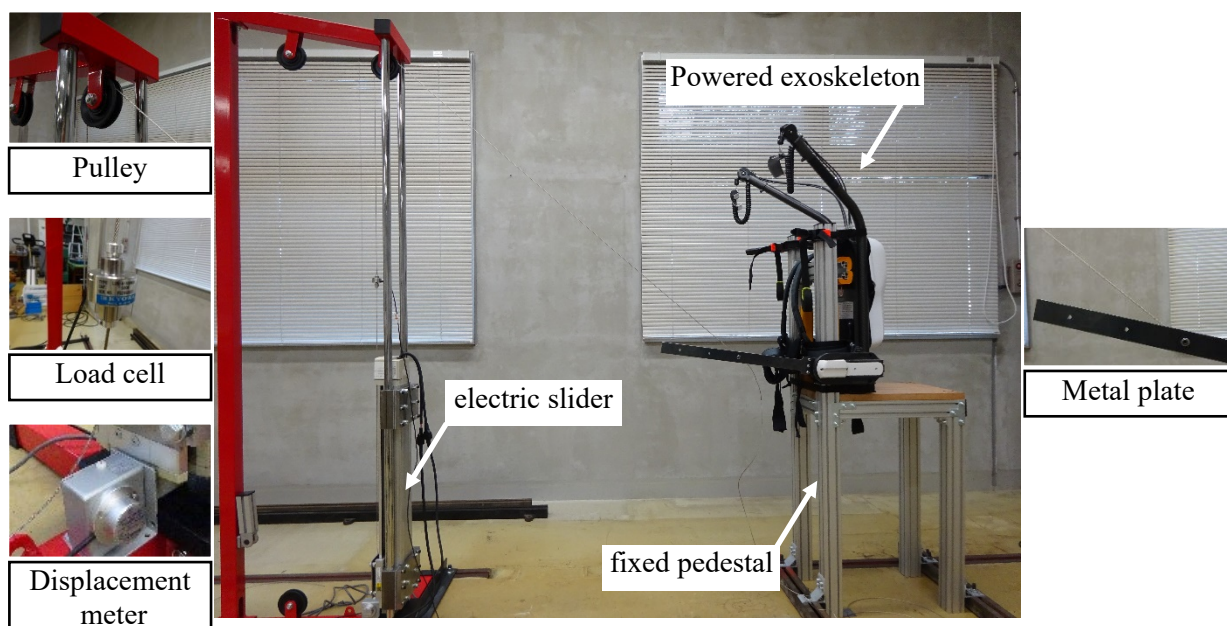**Figure 1. Performance test method regulated in JIS B8456-1**.

Figure 2. Appearance of the measuring instrument we developed

2.2 Safety requirements

We performed detailed check about safety requirements and related item that powered exoskeleton should have satisfy such as risk assessment, construction, shape, size and mass in JIS B 8456-1 while considering the situation where powered exoskeleton was used in agriculture. As a result, most of them were necessary even considering agricultural use and could be applied to agriculture as it was. For example, thought and value of requirements such as "upper limit of assistive torque should not exceed human power ", "powered exoskeleton should have back drivability under working or not working condition" and "protecting users from electric shock" were acceptable and applicable to any industry and field. However, only risk assessment needed to be performed anew in each industry and field that this standard would not assume at the beginning. Therefore, we performed extraction of typical risks expected in agricultural use of powered exoskeleton.

3. RESULTS AND DISCUSSION

The results of our consideration were as follows.

3.1 Performance test method

The result of testing was shown in Fig.3. The characteristics of dynamic assistive torque that the powered exoskeleton has was revealed with high repeatability such as Maximum torque and its duration, torque increasing gradient characteristics and decreasing gradient characteristics were different greatly each step. Maximum and average torque of the powered exoskeleton are following: left side is 44.0 N·m and 40.5 N·m, right side is 47.2 N·m and 41.0 N·m in step of 'high', and 5 times standard deviation of them are lower than 1 N·m.
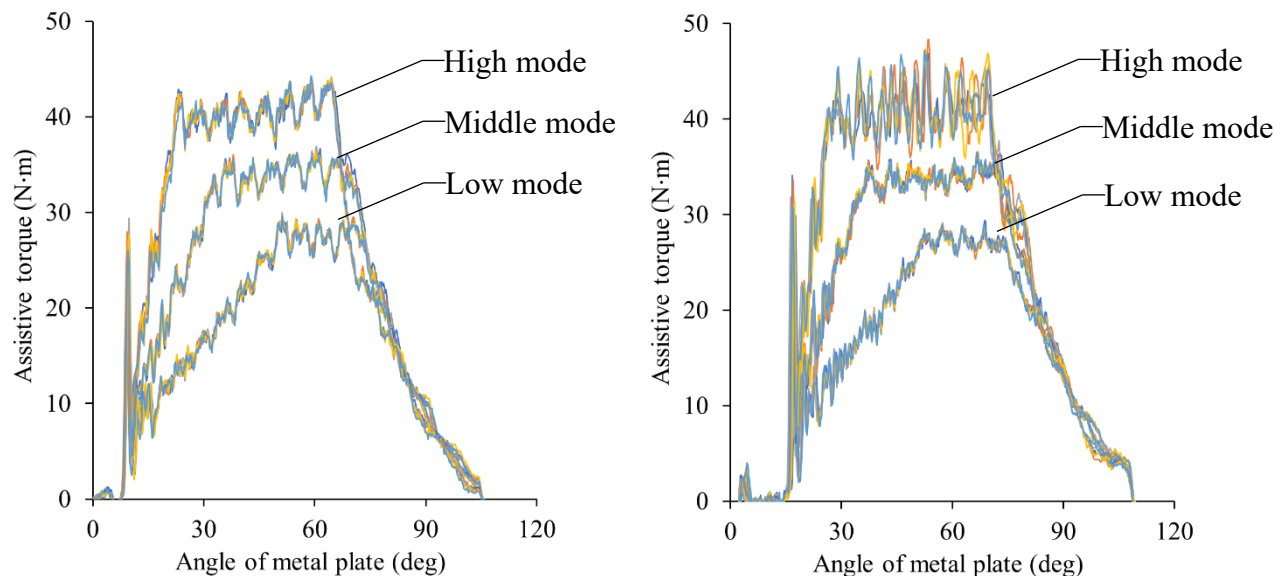
Figure 3. Appearance of the measuring instrument we developed

3.2 Safety requirements

A part of typical risks and the example expected in agricultural use was shown in Table 1. Falling due to unstable ground or slopes such as orchards and field as agricultural specific risks. We considered that detailed examination was necessary what process might lead to falling for user with powered exoskeleton.

Table 1. Typical risks expected in agricultural and the example.

Typical risks expected in agricultural use	Examples
Falling due to unstable ground or poor environment	Work on slopes such as orchards Work in a field with poor footing due to rain etc. Work in heavy rain and wind
Falling due to contact with an object	Contact to the branches of fruit trees in orchards Contact to branches and vines in vegetable cultivation
Obstruction of fall avoidance due to powered exoskeleton working	When use is taken suddenly in the field When use is taken on poor footing or slopes
User's mistake	Overweight User's poor health Lack of user's skill Wrong way to wear

4. CONCLUSION

We considered the applicability of this standard to agriculture and what was necessary for that. As a result, regarding to performance test method, we considered that could be apply to agriculture by using our measuring instrument measuring dynamic assistive torque important in farm work. Regarding safety requirements, risk assessment for agricultural use of powered exoskeleton and detailed examination was necessary such as what process might lead to falling for user with that.

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4:15 PM - 4:30 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D)

[4-1600-D-02] Research on an Intelligent Robot Eye-hand System for Harvesting Pumpkin in the Outdoor Condition

*Liangliang Yang[1], Qian Wang[2], Yohei Hosino[1], Hiroki Ishikuro[1], Ying Cao[1]　(1. Kitami Institute of Technology(Japan), 2. Ning Xia University(China)）
Keywords: Robot, Machine vision, Harvester, Pumpkin

Hokkaido is the largest pumpkin planting region in Japan. However, the planting area is shrinking these years for the shortage of labor force for harvesting the pumpkin fruits. An autonomous robot harvesting system is required by farmers. In addition, the farmers asked us that the surface of the fruits cannot be scrubbed, because of the restrict market requirement of vegetables. Moreover, the storage span will be shorter if the surface has broken points. Therefore, a robot eye-hand system is going to be developed for harvesting the pumpkin fruit. There are three modules of the harvester robot, which are fruits detection module, position coordinates transform and communication module, and robot arm module. In the first module, a color USB camera (IDS, xs, Germany) was utilized to grab images of the field on the speed under 30 frame per second (FPS). The grabbed images were processing in real-time by using faster regional convolutional neural-network (faster R-CNN) method. The pumpkin fruits can be detected correctly around 90% using the method. The detected results were transferred to the second module. In the second module, a PC was connected to the robot arm via an Ethernet cable. The PC was configured as a TCP/IP server and the robot arm was configured as a TCP/IP client. The server and client were communication each other by a speed of 125 Hz, so that the robot arm can be controlled in real-time. The server transformed the position data of the pumpkin fruits from the camera coordinates to the robot coordinates. In the third module, a commercial robot arm (universal robot, UR5, Denmark) was utilized to catch the target fruits.

4:30 PM - 4:45 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D)

[4-1600-D-03] Handy Type Pig Weight Estimation System Based on Random Forest Algorithm

*Hsu Lai Wai[1], Kikuhito Kawasue[1], Khin Dagon Win[1], Kumiko Yoshida[2]　(1. University of Miyazaki(Japan), 2. KOYO Plant Service(Japan)）
Keywords: Xtion-2 Device, Laser Slit, Region Growing, Random Forest, Pig Weight Estimation

In every pig farm, manual pig weight measurement takes time and needs many labors. Generally, load cell is used in pig farms to measure the pig weight. That way of measurement is hard to guide pigs to the weighting machine. The most problem is having vibration when the pig is on the load cell. It causes the inaccuracy result in measuring the pig weight and takes at least 20 seconds to get the stable result. In addition to the pig weight, the labors measure the body length and girth of the pig to know how much changes in pig growth. It is difficult for labors to control the pigs during the measurement. At least two labors are needed to control the pig body. In case of manual measurement, the pig body must be straight to get the stable result since the pig takes different poses. Thus, the mouth of the pig is fixed with the steel wire to avoid pig movement during the pig measurement. That is a hard work for both labors and pigs in every pig farms. In order to cope with these problems, we have developed the handy type measurement system to get parameters to estimate the pig weight by just capturing the image of the pig in the pig farm. The pig body length is defined as the length between the head and tail along the spine of the pig body. The position behind the fore legs of the pigs is known as girth position of the pig. The size of a pig body area is also important in pig weight estimation.

These parameters are extracted automatically by our system, regardless of the posture of the pig.

In our system, Asus Xtion-2 Device is used to estimate pig weight. Laser slit is also used to align the direction of the pig body. Xtion-2 device contains RGB-D sensor and can provide 5M RGB resolution. Thus, clear depth image is captured with that device. That captured image is used as data in the estimation of pig weight.

Therefore, that system not only reduces works and time for labors in the pig farm but also releases pig struggling when guiding the pig to the load cell. After capturing depth image using Xtion-2 device, pig body to be measured is extracted automatically from that captured image. In extraction process of the pig body, Region Growing method is applied in our system. Region growing method can extract the target body robustly from the depth image. After extraction of the pig body, our system detects both 2-D data and 3-D data such as body length and girth to estimate the pig weight.

After extracting the parameters of the pig body of the captured image, Random Forest Algorithm, one of the machine learning method is applied to estimate the pig weight in our system. There are advantages of Random Forest Algorithm. Random Forest can be used for identifying the most important features from the training dataset. Therefore, Random Forest Algorithm is the appropriate method for measuring the pig weight in the practical condition of a pig farm. The estimated pig weight is accurate with the ground truth weight with the use of random forest method. The operator can know the estimated pig weight immediately by just capturing one image for the pig to be measured.

4:45 PM - 5:00 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D)

[4-1600-D-04] Plant Disease Identification using Explainable Features with Deep Convolutional Neural Network

*Harshana Habaragamuwa[1], Yu Oishi[1], Katu Takeya[1], Kenichi Tanaka[1] (1. National Agriculture and Food Research Organization(Japan))
Keywords: Plant disease identification , Explainable features, Convolutional Neural Network , Auto-encoder, Deep learning

Recently deep learning algorithms are widely used in agricultural applications such as disease identification. However, the most of these algorithms are black-box models, which means the users are unable to interpret (explain) what kind of features the Convolutional Neural Network (CNN) algorithm learned to perform the classification task. Without interpreting the learned features, users cannot verify whether the algorithm learned the correct features, this problem may lead to disastrous situations. Because of low interpretability, it is difficult to, improve the training data, gain new knowledge from the data, improve the architecture, or predict the behavior of the algorithm in different conditions. Our objective is to develop a deep learning algorithm which, in an intermediate stage creates explainable features that can be used to discriminate between a healthy and diseased leaf. We used the PlantVillage dataset which is a commonly used dataset for disease identification research, to develop and test our algorithm. This data set consists of leaf images (healthy and diseased) from plants such as tomato, potato, bell pepper, etc. The algorithm is made of three stages. The first stage is an unsupervised generative training using a variational auto-encoder. The second stage involves a supervised generative training using a variational auto-encoder and the final stage involves training a supervised classifier to discriminate between healthy and diseased leaves. The results were evaluated using the visual quality of the features which can be visualized in the second stage of the training. We also tested the final classification accuracy, because there is a compromise between interpretability (understandability) and fidelity (the accuracy of classification). The results of our visual outputs were easy to understand with compared to a conventional heat-map visualization. Our average classification accuracy was

92%, which may be acceptable given the level of interpretation supplied by our method. Our method can be used to find out the features which may be used to separates a healthy and diseased leaf with a low sacrifice to the final classification accuracy. In the agricultural field, this method will help in disease classification to improve algorithms and deficiencies in training datasets. Moreover, the disease experts can predict the behavior of this algorithm in different situations and they can gain knowledge about the features which are characteristics of plant disease. In the future, this algorithm would be extended to other fields where the safety is of paramount importance. Object identification in autonomous vehicles, food safety inspections, and poisonous plant identifications are perspective areas to extend our algorithm.

5:00 PM - 5:15 PM (Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D)

[4-1600-D-05] Sensitivity and Dynamic Analysis of Microalgae Fuel Production System Using LCA

*Riaru ISHIZAKI[1], Ryozo Noguchi[2], Agusta Samodra Putra[1], Tofael Ahamed[2], Makoto M. Watanabe[3]　(1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan), 3. Algae Biomass and Energy System R&D Center, University of Tsukuba(Japan))
Keywords: Bio-production systems engineering, Microalgae, Concurrent engineering, Visualization, Standardization

The purpose of this research is to develop a data analysis system for microalgae that included upstream to downstream processes of production. Energy profit ratio (EPR) and life cycle assessment (LCA) were applied for evaluating production capability refer to output, quality besides output performances, cost, and delivery (QCD). A system approach is required to develop a data analytical platform to increase QCD performances at the different stages of microalgae production based on the concept of concurrent engineering. The forecasting of dynamic result changes could play a key role to data shearing at the different sub-unit of microalgae production system. The microalgae oil-production processes consisted of four sub-unit: open raceway pond (ORP), flocculation, drum filtration, and hydrothermal liquefaction (HTL). The system was based on the experimental data from the Minami-Soma pilot project. Three scenarios were established. Scenario 1 was built a new bioplant on an industrial site to produce a 17.48 kg/day bioclude, by processes algae-containing liquid (50 t/day) from a 0.1 ha ORP. Scenario 2 was built with the thermal power plant site and added the use of heat from there, and the wastewater to scenario 1. In scenario 3, the depth of the water changed from 0.2 to 0.4 meter and related equipment was scaled up to follow scenario 2. The calculated EPR was observed 0.57, 10.81, 10.41 for scenario 1, 2, 3, respectively. The primary contributor was discharged heat from the power plant for utilizing in the HTL process, and replacement of the wastewater treatment energy by microalgae. The EPR was considered from running energy that did not include the energy from the initial investment and input material production. The global warming potential with accumulated value for 100 years (GWP 100) was reported as 47.5, 53.4, 25.4 kg CO_2eq in CO_2 conversion per kg of biocrude for scenario 1, 2, 3, respectively. The acidification potential (AP) and eutrophication potential (EP) had similar trends. In scenario 2, the environmental impact was not changed compared to scenario 1. In this regard, construction of the wastewater treatment plant was added in the system boundaries. In addition, the depth of the ORP at the scenario 3 was doubled. Therefore, the environmental impact per product was observed half compare to scenario 2. The major cost of production was labor and depreciation costs of the HTL plants. Through the LCA-based system approach, microalgal production could be suggested for the best optimal production pattern in any site-specific requirement of environment for sustainability.

Oral Session | Others (including the category of JSAM and SASJ)
[4-1600-D] Other Categories (1)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D (4th room)

[4-1600-D-06] An Aerial Weed Detection System for Green Onion Crops Using the You-Only-Look-Once (YOLO) Deep Learning Algorithm

Addie Ira Borja Parico[1], *Tofael Ahamed[2] (1. College of Agrobiological Resource Sciences, School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan))
Keywords: You Only Look Once, Deep Learning, Weed Detection, Convolutional Neural Network, Unmanned Aerial Vehicle

Herbicide application is a common and inevitable method for preventing weed growth for some crops. Green onions are vulnerable to and significantly affected by weed infestation. However, herbicide contamination can pose as a food safety concern, especially in Japanese cuisine where green onions are typically eaten fresh. As a possible solution, an herbicide spraying system precisely targeting weeds while avoiding green onions was conceptualized. As a preliminary investigation, this study develops and evaluates the performance of what is referred to as the YOLO-WEED, a system that allows the smart detection of weeds through the utilization of unmanned aerial vehicles (UAVs) combined with You-Only-Look-Once (YOLO) deep learning algorithm. YOLO is a forerunner in terms of inference time in object detection, making it suitable for UAV applications. For the dataset, a five-minute UAV video was taken at altitude 4-5 meters at 0-1.3 m/s speed. Each frame from the UAV video were captured and cropped into tiles. 600 of these tiles were selected, annotated and split into training and validation dataset (450) and testing (150). After that, training, validation and testing were performed on YOLO-WEED with the GPU NVIDIA GeForce GTX 1060. IoU, which is the ratio between area of overlap and area of union of the bounding boxes of the ground truth object and the prediction, is the basis of true positive (TP), false positive (FP) and false negative (FN). Based on the TP, FP and FN, the following main performance metrics can be calculated: F1 score (with values 0 to 1) and mean average precision (with values 0 to 100 % with a threshold of 50% for IoU). Moreover, the detection speed expressed in frame per second (FPS) was also determined. YOLO-WEED demonstrated high detection speed (23.7 to 27.8 FPS) and remarkable performance, with mean average precision of 91.09 % and an F1 score of 0.85. YOLO-WEED was also tested on a cropped UAV video and the limitation of YOLO in detecting small objects was minimized. These results successfully show the effectiveness of the YOLO-WEED system for real-time UAV weed detection given its high speed and high accuracy in detection.

An Aerial Weed Detection System for Green Onion Crops Using the You-Only-Look-Once (YOLO) Deep Learning Algorithm

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ABSTRACT

Herbicide application is a common and inevitable method for preventing weed growth for some crops. Green onions are vulnerable to and significantly affected by weed infestation. However, herbicide contamination can pose as a food safety concern, especially in Japanese cuisine where green onions are typically eaten fresh. As a possible solution, an herbicide spraying system precisely targeting weeds while avoiding green onions was conceptualized. As a preliminary investigation, this study develops and evaluates the performance of what is referred to as the YOLO-WEED, a system that allows the smart detection of weeds through the utilization of unmanned aerial vehicles (UAVs) combined with You-Only-Look-Once (YOLO) deep learning algorithm. YOLO is a forerunner in terms of inference time in object detection, making it suitable for UAV applications. For the dataset, a five-minute UAV video was taken at altitude 4-5 meters at 0-1.3 m/s speed. Each frame from the UAV video were captured and cropped into tiles. 600 of these tiles were selected, annotated and split into training and validation dataset (450) and testing (150). After that, training, validation and testing were performed on YOLO-WEED with the GPU NVIDIA GeForce GTX 1060. IoU, which is the ratio between area of overlap and area of union of the bounding boxes of the ground truth object and the prediction, is the basis of true positive (TP), false positive (FP) and false negative (FN). Based on the TP, FP and FN, the following main performance metrics can be calculated: F1 score (with values 0 to 1) and mean average precision (with values 0 to 100 % with a threshold of 50% for IoU). Moreover, the detection speed expressed in frame per second (FPS) was also determined. YOLO-WEED demonstrated high detection speed (23.7 to 27.8 FPS) and remarkable performance, with mean average precision of 91.09 % and an F1 score of 0.85. YOLO-WEED was also tested on a cropped UAV video and the limitation of YOLO in detecting small objects was minimized. These results successfully show the effectiveness of the YOLO-WEED system for real-time UAV weed detection given its high speed and high accuracy in detection.

Keywords: You Only Look Once, Deep learning, Weed detection, Convolutional neural network, Unmanned aerial vehicle

1. INTRODUCTION

Weed control is important for green onion crops as weeds can easily outcompete them because they grow more rapidly, thus, shading the crop and competing for nutrients and water (Gilreath et al., 2008; "Green Onions," 2016; Hewson and Roberts, 1973). The largest expenditures in green onion production in terms of pest management, after all, come from weed control (Norsworthy et al., 2007).

Although elimination of weeds is important, herbicide should not be applied generously to green onions for multiple reasons. One is that herbicide contamination can pose as a food safety concern, especially in Japanese cuisine where green onions are typically eaten fresh. At the agronomy's side, herbicides can

reduce the height, density and yield of green onion when applied during crop emergence in at least 1-2 years (Norsworthy et al., 2007). And from the environment's viewpoint, excessive application of agrochemicals leads to runoff, which can negatively impact the quality of ground water or even contaminate the fisheries. Thus, as much as possible, herbicide application on onion should be minimized. Fortunately, spraying systems can be more precise and improved with deep learning-based weed detection systems.

コメントの追加 [PAIB1]: Add the food safety concern

Machine learning is a set of algorithms that does not need explicit programming to perform a goal by inferencing patterns from input data. Deep learning, on the other hand, is a sub-field of machine learning that is about learning in multiple levels of abstraction in order to model complex relationships among data such as images, sound or text (Deng and Yu, 2014). These layers of non-linear information processing are often called neural networks. In this study, the deep learning algorithm used is YOLO (You-Only-Look-Once), an object detection system that uses a single convolutional neural network, called Darknet, to simultaneously predict the bounding box coordinates and class probabilities straight from an image (Redmon et al., 2015). Its remarkable speed has attracted attention from the deep learning field, boasting a 22 millisecond-inference time with a GeForce GTX Titan X (Redmon and Farhadi, 2018). This makes it highly suitable for real-time applications, such as UAV. However, the cost of increasing the speed is reduced accuracy. Thus, in this study, a YOLO-based weed detection system, which will be called YOLO-WEED from hereon, was developed and evaluated using a UAV video as source of dataset.

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コメントの追加 [PAIB3]: Make this consistent with the abstract

2. MATERIALS AND METHODS

2.1. Data Acquisition
Video acquisition was done in a green onion field with weed incidences in Yatabe, Tsukuba-shi, Ibaraki-ken, Japan (36°00.414'N 140°05.349'E) on a clear day on May 2019 using DJI Phantom 3 Pro. The video was five minutes long recorded at 5-meter altitude in a very slow speed (0-1.3 m/s).

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2.2. Dataset Preparation
The video was divided into image frames by using VLC to capture the video frames per second (one capture for every 24 frames). All screen captures were divided into 96 tiles (8 rows, 12 columns) so that weeds are easier to identify and annotate. Total of 600 images (size 341 x 270 pixels) with both green onion and weeds of differing lighting, clarity, incidence and object sizes were randomly selected. These images were divided into two sets: training & validation (450 images) and testing (150 images). Weeds in the images were labeled with an open-source software BBox-Labeling-Tool (Qiu, 2017). Then, the generated labels were converted into YOLO format. To test YOLO-WEED on videos, a cropped UAV video of resolution 864 x 688 was prepared.

2.3. Weed detection with You-Only-Look-Once (YOLO) algorithm
To set up the weed detection system, Darknet (Redmon, 2016) was downloaded from a stable and improved Github repository of Darknet (AlexeyAB, n.d.). GPU-enabled YOLOv3 (Redmon and Farhadi, 2018) was compiled in Windows 10 operating system using Visual Studio with the dependencies installed (CUDA version 10.1.168, OpenCV 3.4.0, cuDNN 10.1). The hardware used has the following specifications: Quad-core Intel® Core ™ i7-7700HW @ 2.80 GHz, NVIDIA GeForce GTX 1060 and 16GB RAM.

2.3.1. Metrics for evaluating the performance of YOLO
The performance of YOLO was evaluated based on the metrics used in the Pascal VOC Challenge (Everingham et al., 2010). The first metric is Intersection over Union or IoU (equation 1), which is the

ratio between the area of overlap and the area of union of the bounding boxes of the prediction and the ground truth object:

For a detection to be considered as True Positive (TP), or ground truth objects that were correctly identified, IoU should be equal to or greater than 0.5, deliberately set this low to account for human errors in creating the bounding boxes for the ground truth, for example, if a plant has radial stems but most of its vegetative parts are at the center, is somewhat subjective. False positive (FP) detections will be having IoU values under 0.5. False negative (FN) detections are ground truth objects that were completely missed by the predictions or those assigned with low confidences in predictions (eliminated by a certain threshold, which is in this case 0.25).

<div align="center">Table 1. Formulas for the performance metrics used to evaluate YOLO-WEED</div>

Performance Metric	Equation number
$IoU = \dfrac{area\ of\ overlap}{area\ of\ union}$	(equation 1)
$R = \dfrac{TP}{TP + FN}$	(equation 2)
$P = \dfrac{TP}{TP + FP}$	(equation 3)
$F1 = \dfrac{2 \cdot P \cdot R}{P + R}$	(equation 4)
$mAP = \displaystyle\int_0^1 P(R)dR$	(equation 5)

The second metric is Recall (equation 2), or in other words, the sensitivity of the weed detection system. This metric defines the proportion of true positive detections to total ground truth objects. The third metric is Precision (equation 3), which is the proportion of the true positive detections to all positive detections. Next is F1 score, as seen in (equation 4), which quantifies the overall performance of detection by incorporating both precision and recall. Another metric is mean average precision or mAP (equation 5), the area under the precision-recall curve. It is an alternate metric to F1 score in terms of summarizing precision and recall. This metric is often used during the training to select which weights fit the model.

2.3.2. Training YOLO
Default configurations intended specifically for custom object detection for initial training of YOLOv3 (AlexeyAB, n.d.) were set initially with the pre-trained weights for darknet53 (Redmon, 2016). For each 100 iterations, weights were generated during the training process. To determine if training should be halted (to avoid overfitting), the mAPs and loss function (equation 6) chart were enabled and examined. The training was terminated when the average loss no longer decreases as much after many iterations and when the highest mAP has been achieved. For this case, maximum iteration number of 2000 was enough.

$$loss = \lambda_{coord} \sum_{i=0}^{S^2} \sum_{j=0}^{B} \mathbb{1}_{ij}^{obj} \left[(x_i - \hat{x}_i)^2 + (y_i - \hat{y}_i)^2\right]$$

$$+\lambda_{coord} \sum_{i=0}^{S^2} \sum_{j=0}^{B} \mathbb{1}_{ij}^{obj} \left[\left(\sqrt{w_i} - \sqrt{\widehat{w}_i}\right)^2 + \left(\sqrt{h_i} - \sqrt{\widehat{h}_i}\right)^2\right]$$

$$+ \sum_{i=0}^{S^2} \sum_{j=0}^{B} \mathbb{1}_{ij}^{obj} (C - \hat{C}_i)^2 \qquad \text{(equation 6)}$$

$$+ \lambda_{noobj} \sum_{i=0}^{S^2} \sum_{j=0}^{B} \mathbb{1}_{ij}^{noobj} (C - \hat{C}_i)^2$$

$$+ \sum_{i=0}^{S^2} \mathbb{1}_{i}^{obj} \sum_{c \in classes} (p_i(c) - \hat{p}_i(c))^2$$

where $\mathbb{1}_{i}^{obj}$ is when an object appears in cell i and $\mathbb{1}_{ij}^{obj}$ is when the jth bounding box predictor in cell i is "responsible" for that prediction

2.3.3. Validation and testing YOLO

The purpose of validation is to evaluate the performance of the weights generated from the training. Testing is to make sure overfitting is minimized, which means the YOLO-WEED using the generated weights can also detect weeds with other datasets. Validation and testing in this paper was based on the bounding box evaluation of Pascal VOC Challenge (Everingham et al., 2010). Testing was divided into two parts: testing (1) with test images and (2) with the cropped UAV video.

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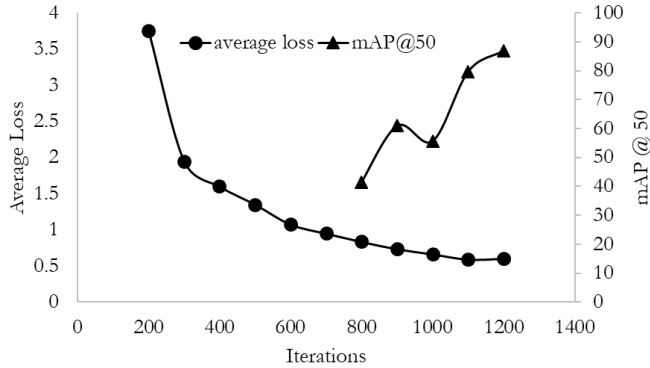
Figure 1. Loss function and mean average precision values (at 50 percent threshold for IoU) during the training at network resolution of 416 x 416, learning rate = 0.001, decay = 0.0005, momentum = 0.0005 and batch size = 64

3. RESULTS

3.1. Training YOLO for weed detection

YOLO was trained with 450 annotated images to detect instances of weeds. Weights were generated every 100 iterations during training. The change in average loss started to become negligible at iteration

1200 so training was stopped at that point. Weights with the lowest loss and highest mAP after the early stopping point (around iteration = 800) were selected for further validation. As seen in *Figure 1*, highest mAPs and lowest losses were achieved at iteration 1100 and 1200.

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3.2. Validating YOLO-WEED

The weights were validated using the performance metrics mentioned in Section 2.3.1. From the training, weights from iteration 1100 and 1200 had the highest performance. And it is confirmed in validation that weights from iteration 1200 may be most suitable for use as characterized by high scores in Average IoU, mAP, Precision, Recall and F1 (refer to *Figure 2*). In addition to this, detection speed at the said iteration was 23.7 FPS, which is deemed enough for standard frame rates of videos (24 FPS).
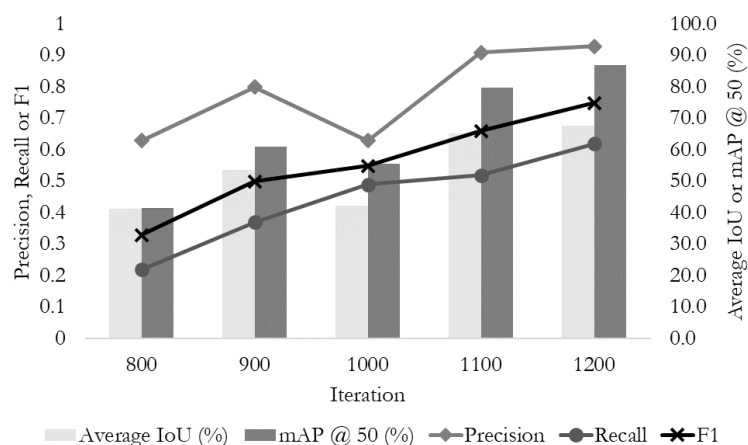
Figure 2. Validation results among weights from iterations on and after the early stopping point. A threshold of 50 percent for the IoU (Intersection over Union) is applied for the mAP (mean average precision).

3.3. Testing YOLO-WEED

Well-performing detection in validation may reflect overfitting so final testing was done with a separate set of 150 annotated images. It is confirmed with high scores in performance metrics that overfitting is minimized at iteration = 1200 (*Table 2*).

In terms of F1-score and mAP @ 50, it can be construed that the weed detection system is well-performing in comparison with YOLO that used official datasets: mAP@50 of 55.3 percent using COCO Dataset (Redmon and Farhadi, 2018) and mAP@50 of 78.6 percent using PASCAL VOC 2007 dataset (Redmon and Farhadi, 2017). However, the YOLO-WEED was only trained with one class, which explains its better metrics compared to YOLO being tested with COCO and PASCAL VOC 2007 datasets.

Table 2. Results from Testing the Trained Network with 150 Test Annotated Images Using Weights from
Iteration = 1200, Network Resolution of 416 X 416

	Value
Average IoU	68.28 %
Precision	0.94
Recall	0.78
F1-score	**0.85**
mAP @ 50*	**91.09 %**

*at 50 percent threshold for IoU

Precision has a very high value of 0.94, which signifies that a remarkable amount of the total positive detections are *true* positive detections. This means that YOLO was able to distinguish green onion from weeds, thus, very little false positive predictions were generated. Recall, however, is relatively lower than precision, which implies that 22 percent of the ground truth objects were not detected at all. This is possibly due to the fact that YOLO is disadvantageous in detecting small objects (Du, 2018). It is also possible that there were not enough weed objects during the training that were occluded, thus, detection of occluded weeds may have lower confidence and were eliminated by the confidence threshold.

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Figure 3. (Top Left) Test image with instance of "weed" object is enclosed in a bounding box of 98 percent confidence at 27.8 FPS. (Top Right) Test image with occluded "weed" object was still detected at 33 percent confidence at 29.4 FPS. (Bottom Left) An image frame from the tested cropped video with instance of "weed" object detected at 88 percent confidence. (Bottom Right) Another image frame from the tested cropped video with smaller weeds detected at relatively lower confidence (28 percent and 38 percent), including a false negative detection.

3.4.   Testing YOLO-WEED on UAV videos

As a final test, the YOLO-WEED was tested on a cropped video (864 x 688 resolution). The detection was done at 512 x 512 network resolution of YOLO. *Figure 3* shows some screen captures of the weed detections, which occurred at approximately 24 FPS. This further proves the high potential of YOLO in

real-time object detection in videos. However, there are still some false negative detections for smaller weeds.

Before testing with the cropped video, YOLO-WEED was first tested on a full-resolution UAV video (4096 x 2160). However, weeds were not successfully detected of which might be due to the spatial constraint of YOLO in detecting extremely small objects. The implementation of the same value for threshold for all object sizes may also contribute to false negative detections. In this paper, a threshold of 0.5 was implemented for IoU. However, small objects' IoU can deviate largely with just slight difference in pixels between the ground truth and prediction (Russakovsky et al., 2015). For example, consider an object of size 10 x 10 pixels with a corresponding detection bounding box of 20 x 20 pixels that fully encloses the object. The IoU in this case will be 0.25, which is eliminated from the threshold of 0.5. Another possible source of problem is the loss function of YOLO, which treats errors the same in all sizes of bounding boxes, big or small. A small error in a large box is generally negligible but a small error in a small box has a bigger effect on the IoU.

4. CONCLUSION

This study developed and evaluated the performance of YOLO-WEED, a system that allows the detection of weeds in UAV video frames through the utilization of the deep learning algorithm YOLO. It demonstrated high speed (23.7 to 27.8 FPS), making it suitable for real-time weed detection for green onion fields. Furthermore, it showed remarkable performance with test images, having a mean average precision of 91.09 percent and an F1 score of 0.85. Lastly, YOLO-WEED performed fairly with the cropped UAV video, having the limitation of YOLO in detecting small objects minimized.

On the other hand, there are still things to improve for YOLO-WEED: the detection of small weeds and the robustness of the detection system. For the first one, training dataset should be collected at lower altitudes (1 to 2 meters) to reduce the need to crop the UAV video, thus, the small weeds will appear larger. This will increase the recall score by preventing false negative detections. To increase the robustness of weed detection, one thing that can be done is to perform data augmentation, of which diversifies the training dataset by generating images (of different scale, viewpoint, rotation, exposure, clarity, resolution and texture) from the same set of original training dataset. Another is to discard training photos that may be considered "difficult" for annotation (Russakovsky et al., 2015) which is often a source of bias in calculation of IoU.

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Oral Session | Others (including the category of JSAM and SASJ)

[4-1600-D] Other Categories (1)

Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D (4th room)

[4-1600-D-07] A Deep Learning and MSM Machine Learning System for Recognition of Weed Infestation in Cabbage Field Using Unmanned Aerial Vehicle

*Tofael Ahamed[1], Yan Zhang[1], Linhuan Zhang[1], Ryozo Noguchi[1]　（1. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan)）

Keywords: Deep Learning , Convolution Neural Network (CNN), Mutual Subspace Method (MSM) , Precision Agriculture, Spot Spraying

Cabbage is susceptible to grow due to weeds incidence and requires large amounts of herbicides in the small-scale Japanese farms. In precision application of herbicides, it is required to recognize the classifiers to minimize herbicides application. Therefore, the purpose of this research is to deal with recognition of weed infestation in the cabbage field using two classifiers: cabbage and weeds. A DJI Phantom UAV was flown with an onboard 4K RGB camera from 2m heights to identify the weed incidence in a cabbage field located at the Ibaraki Prefecture of Japan. Two videos were used and converted to figures: one for training and other for testing. In the pre-process stage, each original image with size of 1920x1080 was divided into 250x250 small size sub-graphs using a sliding window, with step of 250. Each sub-image could be defined as: cabbage and weeds. In the training, 676 datasets for cabbage, 667 datasets for weeds were taken from sub-images. Alexnet CNN deep learning and Mutual Subspace Method (MSM) machine learning were used to find the recognition of the two classes.The accuracy for recognizing the classifiers using MSM was 61%. To improve the accuracy of MSM, Histogram Oriented Gradient (HOG) method was used with MSM. The recognition accuracy was increased to 88% using MSM-HOG algorithm. The overall accuracy was achieved 94%for recognizing the classifiers using AlexNet CNN.In the deep learning process, the enlarging dataset and learning technology can be inherit with AlexNet CNN and MSM-HOG. Further research will be carried out using weights in each of the layer to improve the accuracy of the classifiers. This deep-learning approach has the potential to add in the spot sprayer for real-time application to minimize herbicides for UAV.

A Deep Learning and MSM Machine Learning System for Recognition of Weed Infestation in Cabbage Field Using Unmanned Aerial Vehicle

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ABSTRACT

Cabbage is susceptible to grow due to weeds incidence and requires large amounts of herbicides in the small-scale Japanese farms. In precision application of herbicides, it is required to know classifiers for spot spraying to minimize herbicides application for enabling GAP practices. The purpose of this research is to deal with recognition of weed infestation in the agricultural field, especially for cabbage with two classifiers: cabbage and weeds. Alexnet CNN deep learning and Mutual Subspace Methods (MSM) were used to find out the recognition of the classes. The overall accuracy was achieved 94% for recognizing the classifiers. Further research will be carried out using weights in each of the layer to improve the algorithms and accuracy of the classifiers.

Keywords: Deep Learning, CNN, MSM, Precision Agriculture, Spot Spraying

1. INTRODUCTION

Most of the weeds classification techniques, which are referred to the ground carrier, and the complex image processing algorithms are used for feature extraction and then classified. This makes the method with poor adaptability and lack of robustness. Convolutional Neural Network (CNN) has the potential for recognizing classifiers. On the other hand, MSM has the robustness with fastest computational time for recognizing classifiers (Yan et al. 2018). The CNN has made a rapid development since its appearance in the field of deep learning. It has shown excellent performance using MSM and HOG for shorter time in the field of image recognition, target location and detection. At present, CNN has been studied and applied in many fields (Zheng et al. 2017, Zhao et al. 2018). The well-known AlexNet CNN architecture can be utilized in combination with a sliding window object proposal technique. In this paper, a low-altitude cabbage images were captured by UAV. The CNN and MSM methods are proposed for weed classification considering density of weed infestation.

2. MATERIALS AND METHODS

2.1 Convolution Neural Network (CNN)

CNN is a multi-layer data processing algorithm, which is originally inspired by neural mechanisms underlying visual system and was designed in view of the two-dimensional shape's identification. The convolution layer is the core part of the convolutional neural network. Its main function is to extract local features of the input through the fixed-step movement of the convolution kernel. The core of the convolutional layer operation is to reduce unnecessary weight connections, introduce sparse or partial connections, and bring the weight sharing strategy to reduce the number of parameters greatly. The most common mathematical expression of convolutional layer is as follows:

$$x_j^n = f\left(\sum_{i \in M_i} x_i^{n-1} * k_{ij} + b_j^n \right) \tag{1}$$

where x_j^n represents the j th feature map of the n th convolutional layer, $f(.)$ represents the activation function, M_i represents the selected input feature map combination, x_i^{n-1} represents the i th output feature of the $n-1$ th layer, and "*" represents the convolutional operation, k_{ij} represents the convolution kernel between the i th feature map of the previous layer and the j th feature map of the current layer, and b_j^n is the bias of the current layer. The pooling layer obtains the invariant properties of the higher level by the function transformation of the non-overlapping rectangular region on the upper output characteristic graph. The mathematical expression of the pooling layer is as follows:

$$x_j^n = f(\beta_j^n * down(x_j^{n-1}) + b_j^n) \qquad (2)$$

Where x_j^n represents the j th feature map of the n th pooling layer, $f(.)$ represents the activation function, $down$ represents the pooling process, and β_j^n is a multiplicative weight value, the general value of 1; b_j^n is additive bias, the general value of 0 matrix.

2.2 Mutual Subspace Method (MSM) and HOG

MSM is the extension of the Subspace Method (SM). Classifying a set of input pattern vectors into several classes based on multiple canonical angles between the input subspace and class subspaces (Figure 1. a). HOG is an edge orientation histograms based on the orientation of the gradient in localized region that is called cells. It is easy to express the rough shape of the object and is robust to variations in geometry and illumination changes with MSM (Figure 1. b).
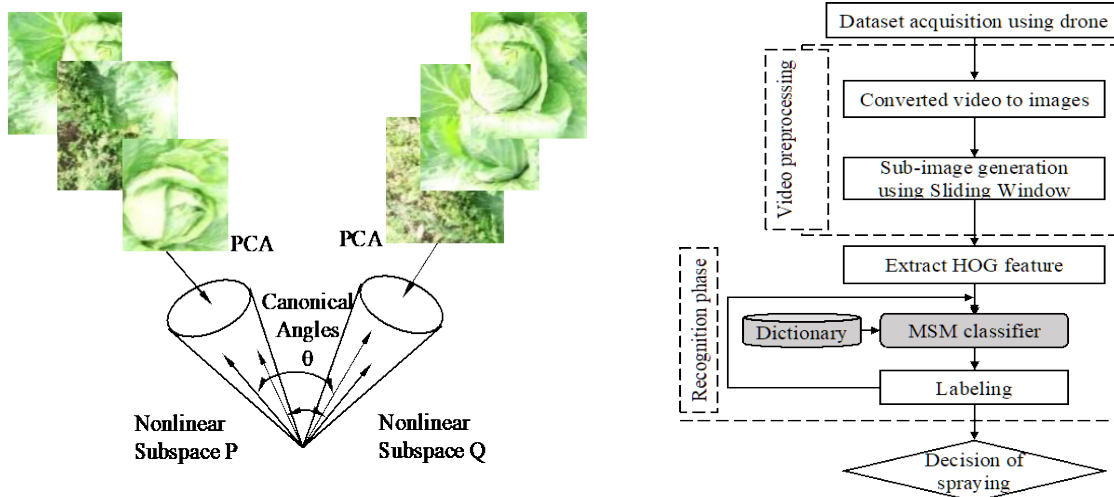
Figure 1 a) Two sets of images using MSM b) Online recognition: HOG & MSM classifiers

2.2 Data Acquisition System

In the cabbage filed, A DJI Phathomon UAV was flown with an onboard 4K RGB camera from 2m heights. From the view of uses, the cabbage is with high production and it's a leafy vegetable, the precision spray for broadleaf type vegetables is more meaningful than others. Two videos were used and converted to figures: one for training and other for testing.

Figure 2 Classes for training and testing for weeds and cabbage including sliding window for sub-images in the original image

The data model AlexNet was used. In the pre-process stage, each original image with size of 1920x1080 was divided into 250x250 small size sub-graphs with a sliding window, with step of 250. Each sub-image can be defined as: cabbage and weeds (Figure 2). In the training, 676 datasets for cabbage, 667 datasets for weeds were taken from sub-images (Table 1).

Table 1 Date sets (data numbers) used for training and testing:

Category	Original image	Sub-images	
		Cabbage	Weeds
Training	136	676	667
Testing	132	303	369

3. RESULTS AND DISCUSSION

The accuracy for recognizing the classifiers using MSM was 61% and 88% using MSM and MSM-HOG (Figure 3. a and b). The overall accuracy was achieved 94% for recognizing the classifiers using AlexNet CNN (Figure 3. c and d). Further researches need to be carried to increase the classifiers for UAV-based autonomous sprayer development. Real-time recognize weeds density level using deep learning model. In the deep learning process, the enlarging dataset and learning technology can be inherit with AlexNet CNN and MSM-HOG.
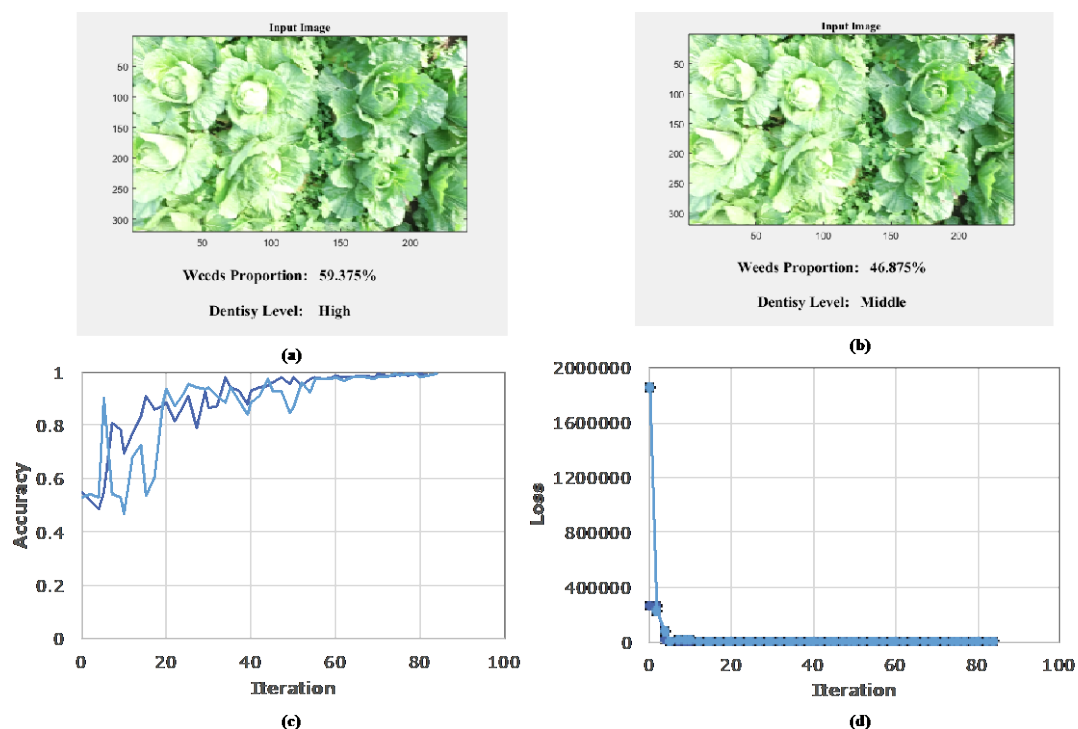
Figure 3 (a and b) MSM and Hog-based accuracy relates weed density level (c and d). Accuracy and loss value in different iterations during training of datasets

In further research, in each of the 250x250 small size image, weight will be set for each class: graphs belong to class of cabbage with weight 1; graphs belong to class weeds with weight 0.5. Finally, a total weight can be calculated and the density level can be judged on-line by comparing a level standard.

4. CONCLUSION

The low-altitude images were taken from a UAV for classification of weeds using AlexNet CNN and MSM-HOG. This deep-learning approach has the potential to add in the spot sprayer for real-time application to minimize herbicides from UAV sprayers. The machine learning systems and inheritance knowledge could help in development of autonomous UAV-sprayer.

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Oral Session | Others (including the category of JSAM and SASJ)
[4-1600-D] Other Categories (1)
Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D (4th room)

[4-1600-D-08] Mallard Navigation Using Unmanned Ground Vehicles, Imprinting, and Feeding

*Hirokazu Madokoro[1], Satoshi Yamamoto[1], Hanwool Woo[1], Kazuhito Sato[1] （1. Akita Prefectural University(Japan)）
Keywords: rice-duck farming, feeding, imprinting, mallards, navigation, unmanned ground vehicle

This study was conducted to develop an unmanned ground vehicle (UGV) that navigates mallards to achieve high-efficiency rice–duck farming. This paper presents two navigation approaches and fundamental experiments to test them using a UGV. As the first approach, we used imprinting applied to baby mallards. Baby mallards, after hatching, exhibit following behavior to a UGV after imprinting. One week after hatching, baby mallards show wariness against an imprinted target object. Experimentally obtained observation results revealed the importance of providing imprinting immediately after hatching. As the second approach, we used feed placed on the top UGV body. Experimentally obtained results showed that adult mallards exhibited wariness not only against the UGV, but also against the feed box. After relieving wariness with provision of more than one week time to become accustomed, adult mallards ate feed in the box on the UGV. However, they ran away immediately at a slight movement of the UGV. We confirmed the necessity of imprinting to baby mallards for actualizing navigation because adult mallards are alerted by the UGV.

Mallard Navigation Using Unmanned Ground Vehicles, Imprinting, and Feeding

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ABSTRACT

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Keywords: rice-duck farming, feeding, imprinting, mallards, navigation, unmanned ground vehicle

1. INTRODUCTION

Rice-duck farming is an environmentally friendly rice cultivation method that employs neither chemical fertilizers nor pesticides. Although hybrid ducks are generally used for rice-duck farming, farmers in northern Japan use mallards because of their utility value as a livestock product. For this study, we specifically examine rice-duck farming using mallards considering regional characteristics. Fig. 1 depicts rice-duck farming using mallards.

Since ancient times, mallards have been domesticated as poultry for human consumption. Mallards are used not only for rice-duck farming, but also for meat because their smell is not strong. Mallards eat leaves, stems, seeds, and shells of plants. For weeding and pest control, mallards eat not only aquatic weeds such as itEchinochloaesculenta, itCyperusmicroiria, and itJuncuseffuses, but also aquatic insects such as itLissorhoptrusoryzophilus, itSogatellafurcifera, itNilaparvatalugens, itLaodelphaxstriatellus, but also river snails in a paddy field. Mallard movements in a paddy field also produce positive effects of full-time paddling. For weed prevention, turbid water suppresses photosynthesis of weeds below the surface. Moreover, mallards not only provide feces for nutrients of growing rice, but also contact rice during movements as a stimulus. A paddy field is a place of abundant water and life for mallards [1]. Moreover, the grown paddy rice can provide refuge from natural enemies.

This study was conducted to develop an unmanned ground vehicle (UGV) that navigates mallards to achieve high-efficiency rice-duck farming. As a navigation method used for mallards, we examined three approaches: imprinting, pheromone, and feeding. This paper presents basic navigation experiments of imprinting and feeding using a UGV.

Figure 1: Rice-duck farming using mallards.

Figure 2: Stepping pond: paddy rice is eaten by mallards.

2. NAVIGATION METHODS

In a paddy field, mallards often concentrate in a specific area because of a swarm habit. Some areas therefore have persistent weeds because mallards do not disperse. Moreover, a stepping pond oc-curs, as depicted in Fig. 2. Mallards eat paddy rice if weeds are insufficient. Moreover, as depicted in Fig. 2, a stepping pond occurs where all the paddy rice has been eaten by mallards. Mallards do not weed a whole paddy field uniformly. Therefore, farmers must weed them using a weed removing machine. As a different approach, some farmers use feed to navigate mallards. The difficulty of this approach is the necessity of human burdens, especially for large paddy fields. For this study, we examined three navigation approaches used for mallards: imprinting, pheromone tracking, and feeding.

Imprinting is a unique behavior observed in nidifugous birds such as ducks, geese, and chickens [2]. Imprinting is a contacting and following response to a stimulus that is received for the first time during a short period after hatching. Moreover, imprinting is enhanced for running and following in response to sounds or moving shadows. For this study, we specifically examined imprinting-based navigation. We used a UGV as an imprinting target for baby mallards.

Pheromones are chemicals that promote changes in behavior and development of conspecific individuals after being produced inside the body and secreted outside the body [3]. Pheromones are used mainly when insects communicate with conspecific individuals. Although no pheromone is available to navigate mallards, we consider indirect navigation using insects favored by mallards with pheromone. Specifically, we devised an indirect usage that navigates mallards using insects that are gathered to a pheromone trap attached to a UGV. Although baby mallards attempt to eat insects, adult birds have no interest in them. We consider that the efficiency of this approach decreases along with mallard development. For this study, we conducted no experiments using pheromone-based navigation because of the difficulty of the procedures using insects and the weak overall effects.

For rice-duck farming, breeders use feed to collect ducks. Breeders give minimum feed for ducks because ducks stop eating weeds if too much feed is given. We expect that feeding-based navigation is effective for adult mallards because imprinting can only be performed during the baby mallard period. Yamada et al. tested the effects of feed learning and navigation for hybrid ducks. However, they described no test for feed learning for mallard navigation. For this study, we specifically examined the method of navigating mallards following the use of feed combined with a UGV.

Figure 3: Inside (left) and outside (right) of the mallard farm.

Figure 4: Mallard eggs (left) and incubator (right).

3. IMPRINTING-BASED NAVIGATION

3.1 Experiment setup

For this experiment, we attempted imprinting of baby mallards after a hatch. We obtained eggs from a mallard farmer. Fig. 3 depicts the outside and inside of the mallard farm.

Figure 4 depicts the mallard eggs and an incubator (MX-20; Autoelex Co. Ltd.) that we used for a hatch. We divided 18 eggs into four groups because we shifted the hatching date in two-day intervals. We set the temperature and humidity in the incubator respectively to 37.5 deg and 45.0%. The bottom incubator plate includes a slider to roll the eggs periodically. We set the rolling frequency to one-hour intervals. We stopped the rolling approximately one week before the predicted hatching date. Simultaneously, the humidity was increased to 65%. The left panel of Fig. 5 depicts a baby mallard immediately after hatching. We kept the baby mallards in the incubator until their feathers were dry. After drying, we moved them to a breeding cage, as depicted in Fig. 5 right. We kept them warm in the cage using an electric heater.

The left panel of Fig. 6 depicts a 3D mallard model used for imprinting. After creating the model using a 3D printer, we put it on the UGV as depicted in the right panel of Fig. 6. The model was made of acrylonitrile butadiene styrene.

Figure 5: Baby mallard after hatching in incubator (left) and moving to a cage after drying of its feathers (right).

Figure 6: 3D mallard model (left) and mounting on UGV (right).

3.2 Experiment results

We applied imprinting sequentially to three mallards. We manually moved the UGV in the cage during 3 hr in front of a baby mallard. After imprinting, the baby mallard attempted to follow the UGV. Subsequently, we observed that the baby mallard changed responses of calling patterns according to UGV movements. The baby mallard moved near the electric heater when the UGV stopped.

We applied imprinting to the second baby mallard, which hatched two days later, with similar stimulation patterns and procedures. Although the second baby mallard followed the UGV, both were interested in each other. Our observations suggest that the interest between them was higher than that for the UGV. Therefore, we put a partition in the cage to separate them. First, they attempted to cross the partition. We showed the UGV to each baby mallard in their separated zone for enhanced imprinting. They continued the following behavior against the UGV. Subsequently, we added the third baby mallard to the cage after installing a partition. The imprinting procedure for the third baby mallard was similar to that used for the former two baby mallards.

For observing the following behavior, we moved the three baby mallards and the UGV from the cage to the test file with the size of 2.0 m2. Fig. 7 depicts time-series images of this experiment. The four sides of the test field were surrounded by wooden boards. As an objective after imprinting, the baby mallards followed a UGV that moved slowly. They continued following the UGV after increase of the velocity.

Imprinting among baby mallards after hatching occurred in a cage because they moved frequently. Based on the experiment for baby hybrid ducks conducted by Yamada et al. [4, 5], we considered that imprinting was enhanced for separating baby mallards because they spent time longer with the UGV together. Moreover, imprinting of individuals was effective because the calling sound volume of separated baby mallards was greater than that of the gathered baby mallards.

Figure 7: Following behavior displayed after imprinting.

Figure 8: UGV used for this experiment.

We attempted imprinting to baby mallards at one week after hatching using a similar procedure with the UGV. We confirmed that baby mallards falsely recognized the UGV as an enemy. Moreover, we observed that they avoided the UGV with a big call. Actually, mallards reach a peak sensitive period at 15 hr after hatching. Subsequently, the imprinting effect is reduced considerably. For the UGV, the interest for baby mallards arose at more than one week after hatching because the socalled critical period is weaker than that of baby mallards several hours after hatching. Experimentally obtained results reveal that navigation is possible for baby mallards before the critical period of imprinting.

4. FEEDING-BASED NAVIGATION

4.1 Experiment setup

For this experiment, we conducted navigation experiments for adult mallards using feed and another UGV. Fig. 8 depicts the UGV (Blizzard FR1/12EP Belt Vehicle; Kyosho Corp.) after attaching a feed box (FB). The body is 488 mm long, 320 mm wide, and 220 mm high. We installed a water-protected monocular camera (CS-QR300; Planex Communications Inc.) at the UGV rear top. The image resolution was one million pixels.

Figure 9: Exterior (upper) and inside (lower) of the aviary.
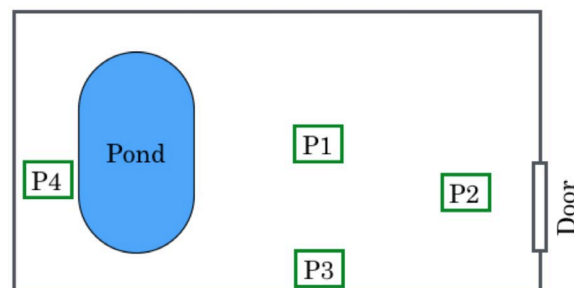
Figure 10: Layout and assignment of the UGV and FB.

4.2 Experiment results

For this experiment, we used nine adult mallards (six months old) without imprinting. We conducted feed learning experiments for them, not only getting used to the UGV while eating feed, but also following the UGV with a desire to get feed. To relieve wariness, we changed the ten experiment conditions in terms of positional combinations between the FB and the UGV, feed types, and sound stimulation. Mainly, we used compound feed that comprises corn, soybean, milo, bran, press cake, and fish flour. Table 1 denotes the experimental conditions including dates and hours.

For the first experiment, we placed the UGV and the FB at P1. We played a sound clip of calling recorded in advance using a microphone. Figure 11 depicts images obtained using the camera on the UGV. The mallards gathered at a corner in the aviary. They did not attempt to approach the UGV because they might have alerted. We observed that they seemed to watch the UGV while keeping their distance. As described herein, we gave no feed to the mallards from the morning of the experiment day. Although they were hungry, they did not approach the feed because of their own wariness. Compared with curious baby mallards, adult mallards were more vigilant against something that they saw first. We considered that the fear of natural enemies was attained by adult mallards for their work in paddy fields.

Table 1: Experiment conditions

No.	Date	Time	UGV	FB
1	Nov. 30th	13:00–14:00	P1	P1
2	Dec. 4th	11:00–13:00	P1	P1
3	Dec. 4th	13:00–13:20	P2	P1
4	Dec. 4th	13:20–13:30	P2	P3
5	Dec. 4th	13:30–13:40	–	P3
6	Dec. 4th	13:40–13:50	P4	P3
7	Dec. 12th	13:30–14:30	P3	P3
8	Dec. 14th	13:00–14:00	P3	P3

Figure 11: Behavior and response patterns for the first experiment.

One week later, we conducted the second experiment. We gave no feed to the mallards during the two days prior. The UGV and the FB were placed in a similar position with playing of a similar sound clip. As additional feed, we gave them cabbage and broccoli, which are vegetables preferred by mallards. The mallards gathered to a corner in the aviary. They showed no approach to the FB on the UGV. After a few minutes, we observed that their wariness had disappeared. Two mallards watched the UGV from the pond. However, they showed no approach to the UGV.

For the third experiment, the UGV position was   changed to P2 near the entrance whereas the FB position remained at P1. We expected that mallards would eat the feed irrespective of wariness of the UGV because the UGV and FB were located separately. However, they did not approach the FB.

For the fourth experiment, the FB position was changed to P3, although the UGV position remained at P2. Moreover, we changed the feed to rice with small granularity. Mallards like to eat rice more than compound feed. However, they showed no approach to the rice.

For the fifth experiment, we removed the UGV, although the FB position remained at P3. Although the UGV did not exist in their view range, they exhibited no approach to the FB.

For the sixth experiment, we placed the UGV at P4 whereas the FB position remained at P3. During the previous five experiments, mallards frequently gathered P4 and its surrounding area. For this experiment, mallards gathered at a position between P3 and P4. Particularly, they moved near P3 when the UGV moved P4. However, they showed no feed eating.

The seventh experiment was conducted eight days after the sixth experiment. We set this period for mallards that became accustomed to the FB. The UGV and the FB were placed at P3. The one-hour observation result demonstrated that mallards showed no feed eating.

Before the eighth experiment, we left the UGV for two days in the aviary. The UGV and the FB were placed at P3, which was a similar position to that used in the previous experiment. As an experimentally obtained result, Fig. 12 depicts images obtained from the camera on the UGV. Mallards were swimming in the pond after relieving wariness, as depicted in the upper two images. Twenty minutes later, one mallard approached the UGV with feed, as depicted in the bottom two images of Fig. 12. Other mallards gathered around the UGV to eat the feed. We moved the UGV slightly while they were eating. They ran away immediately when the UGV moved. As described herein, they approached the UGV from rear. We considered that this behavior derived from a rapid escape response to a dangerous situation.

Figure 13: Breeder to give feed to ducks.

After stopping the UGV, we attempted to approach the mallards. Fifteen minutes later, another mallard approached. The mallard gradually shortened the distance to the FB. Other mallards were eating feed. We forwarded the UGV slightly again. Although the mallards fled all at once, the escape distance was shorter than that of the UGV movement. Apparently, the wariness was relieved for mallards. Although mallards approached from a large place behind, they gathered for eating feed from a narrow direction behind.

4.3 Discussion
Results obtained from eight experiments indicate that adult mallards had strong wariness. We demonstrated that at least two days are required to get used to the UGV and the FB. Moreover, they do not always eat feed. Maintaining navigation using feed alone is expected to be difficult because mallards eat feed only several times a day. As described herein, a breeder gave feed with similar behavior from a similar place every day, as depicted in Fig. 13. We infer that mallards recognize the input combined with breeder's behavior patterns. For this experiment, the FB was placed in the aviary with the feed placed in advance. We infer that the similar behavior to enter feed to a vacant FB on the UGV contributes to wariness relaxation for mallards. Moreover, we infer that the wariness for the UGV movements is relieved if they recognize it as a harmless object.

5. CONCLUSION

To actualize highly efficient rice-duck farming, this study presented experimentally obtained results to verify a useful method to navigate mallards using the UGV combined with imprinting and feeding. Experimentally obtained results revealed that baby mallards with imprinting followed the UGV. We considered that adult mallards require more than two days to become accustomed to the UGV and the

FB because they have strong wariness against unknown objects. Although we did not actualize mallard navigation, we found that mallards ate feed in the FB on the UGV. We confirmed the necessity of conducting imprinting during a baby term for mallard navigation.

As future work, we expect to quantify the imprinting effects of shared time between the UGV and mallards in a breeding process. Moreover, we expect to conduct a navigation experiment using feed of several types in a paddy field for baby mallards after imprinting.

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Oral Session | Others (including the category of JSAM and SASJ)

[4-1600-D] Other Categories (1)

Wed. Sep 4, 2019 4:00 PM - 6:15 PM Room D (4th room)

[4-1600-D-09] Onion Bulb Counting in a Large-scale Field Using a Drone with RTK-GNSS

*Satoshi Yamamoto[1], Hirokazu Madokoro[1], Yo Nishimura[1], Yukio Yaji[1] （1. Akita Prefectural University(Japan)）

Keywords: Onion, Drone, RTK-GNSS, Orthomosaic, Deep learning

RTK-GNSS mounted on a drone made it possible that relatively accurate orthomosaic and digital elevation model (DEM) were generated with minimum number of ground control point (GCP) in a large-scale onion field. Deep learning was then applied to detect onion bulbs in the orthomosaic images. We annotated the bulbs of 16,812 with 250 images for the object detection. After machine learning process, the detection rate was 0.60 while the recall was 0.16. When the trained model was applied to the orthomosaic images, the number of the onion bulb ranged from 29,970 to 109,694 which were from 5 % to 19 % of the estimated onion plant number. Our next step would be to improve the trained model by reducing the false-negative value in the confusion matrix.

Onion Bulb Counting in a Large-scale Field Using a Drone with RTK-GNSS

Satoshi Yamamoto[1*], Hirokazu Madokoro[2], Yo Nishimura[1], Yukio Yaji[1]

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ABSTRACT

RTK-GNSS mounted on a drone made it possible that relatively accurate orthomosaic and digital elevation model (DEM) were generated with minimum number of ground control point (GCP) in a large-scale onion field. Deep learning was then applied to detect onion bulbs in the orthomosaic images. We annotated the bulbs of 16,812 with 250 images for the object detection. After machine learning process, the detection rate was 0.60 while the recall was 0.16. When the trained model was applied to the orthomosaic images, the number of the onion bulb ranged from 29,970 to 109,694 which were from 5 % to 19 % of the estimated onion plant number. Our next step would be to improve the trained model by reducing the false-negative value in the confusion matrix.

Keywords: onion bulb, growth measurement, drone, RTK-GNSS, orthomosaic, deep learning

1. INTRODUCTION

Onion (*Allium cepa L.*) is one of the staple vegetables in Japan. More than 60 % of onion is produced in Hokkaido, large northern islands of the country. Other main producing areas are in south part. In Japanese onion production, off-harvest season is in July and August. The market price is relatively high during the period. Ogata village is well known as a large-scale rice production area for more than 50 years. Recently, some farmers have started onion production in the region for more stable management because the harvest season of onion is from June to July in Akita prefecture. Monitoring crops throughout the vegetation period is essential to achieve higher yield and quality in onion production. However, it should be difficult to check the health of plants accurately in a large-scale field by human-eyes. Drone, which can move around all the field automatically, seems to be a useful tool to scout crops grown in large field. To construct accurate three-dimensional model of the field, many ground control points (GCPs) are necessary because the positioning sensor of a usual drone has an error ranging up to 10 m. It is tedious work to correct camera alignment referring GCPs by human eyes. Furthermore, it is difficult to set GCPs in the middle of agricultural field because it should be obstacles for a tractor and other farm machineries. If a drone can obtain accurate position information, it is thought that the number of GCP could be reduced dramatically. To generate accurate 3D model, LiDAR (Christiansen et al., 2017) and RTK-GNSS (Pix4D, 2017, Chen, 2017) have been researched. RTK-GNSS is a kind of position measuring device which brings extremely precise position information. Even if high precision orthomosaic of the onion field applying a drone with RTK-GNSS and least number of GCPs could be generated, it should be difficult to get valuable information without analyzing the orthomosaic in a stable manner. Deep learning seemed to be one of strong method to detect the target robustly.

In this study, our objective is to clarify how to conduct crop monitoring efficiently and accurately. Onion bulb counting was thought to be a suitable operation to test the developed monitoring system.

2. MATERIALS AND METHODS

2.1 Measurement system

A digital single lens reflex camera is ideal high-performance camera to obtain plant growth information from points of optical quality and data quantity. Figure 1 shows components of the measurement system. As a drone which can carry the heavy measuring equipment, we used Matrice 600Pro from DJI which can lift up weight of 2.6 kg and the typical flying time is about 30 minutes. A digital single lens reflex camera EOS 6D Mark II from Canon was mounted on a gimbal Ronin MX from DJI. The image size was 6240 x 4160. To capture an image by remote control, infrared light system was set in front of the sensor of the camera. The operator can check the image with iPad which is attached to remote controller because HDMI signal is sent from camera to iPad through the drone system. Basically, the camera and the drone doesn't communicate each other only sending one-way signal.
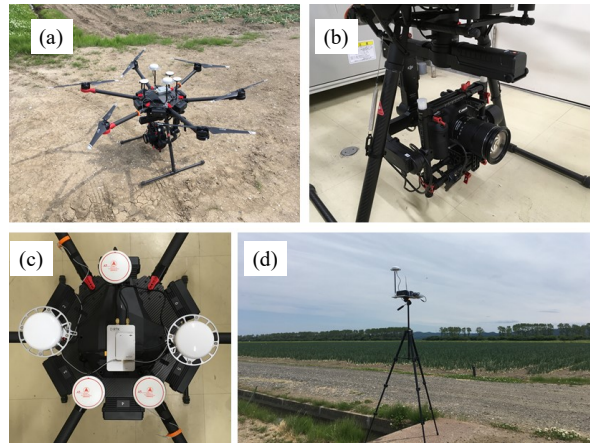
Figure 1. Drone components: (a) Matrice 600Pro from DJI. (b) a digital single lens reflex camera: EOS 6D Mark II from Canon, and a gimbal of Ronin MX from DJI. (c) two RTK-GNSS receivers and three redundant GNSS receivers. (d) a base station for RTK-GNSS .

The original drone has three redundant GNSS antennas to improve the robustness of position measurement. But the accuracy is approximately less than 10 m without correction signal from base station. RTK-GNSS brings positioning accuracy of less than 5 cm. Base station of RTK-GNSS is necessary to achieve the accuracy. Figure 2 shows trajectories of the drone.
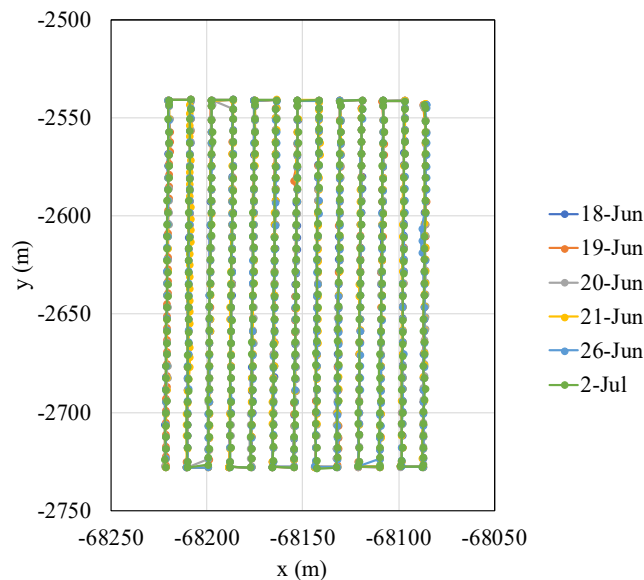
Figure 2. Flight trajectories on June 18th, 19th, 20th, 21st, 26th and July 2nd.

The data was recorded in drone memory during flights. In six flights, the routes are almost same when the RTK-GNSS receiver mode was fix. But the RTK data wasn't written to EXIF of the image because the camera and the drone weren't linked. If a drone stops when grab an image, the position data should be accurate. However, if the images are captured while the drone moves, the position data should have some errors. The camera had its own GNSS receiver, and the GNSS time was recorded to EXIF. According to the GNSS time of EXIF, nearest two of GNSS time of RTK data were selected, then position was calculated based on the data. They are processed with 3D reconstruction software Metashape from Agisoft. When images were grabbed in the large-scale onion field, the number of images was 386 to 430. In Metashape, workflow was as follows: loading photos into the software, aligning photos, building dense point cloud, building digital elevation model (DEM), building orthomosaic, and finally, exporting results. The resolution of the orthomosaic and DEM is about 7 mm/pix and 3 cm/pix respectively. As minimum preparation by hand, three GCPs were set for each data set.

2.2 Onion bulb detection

From the orthomosaic, onion bulbs are observed to be counted by image processing. To detect the onion bulb robustly, deep learning was then applied to generated orthomosaic images. For the machine learning, image size should be 320 x 192. So we separated the orthomosaic into approximately 9000 small images, then, 250 images are randomly selected, then we annotated the bulbs of 16,812 manually. Annotation is relatively easy because the onion bulbs were located in line as shown in figure 3. HALCON, image processing software, provides deep learning function with prepared as compact type using "object detection". The detection rate (TP/(TP+FP)) was 0.6, the recall (TP/(TP+FN)) was 0.16 with intersection over union (IoU) was 0.5. The field size was 2.4 ha; 135 m x 180 m. Four rows distance was 0.24 m, plant distance was 0.1 m, the number of plant bed was 107 rows in field width 180 m. The length of row was 132 m, the number of onion plants in the field was estimated to 564,960.
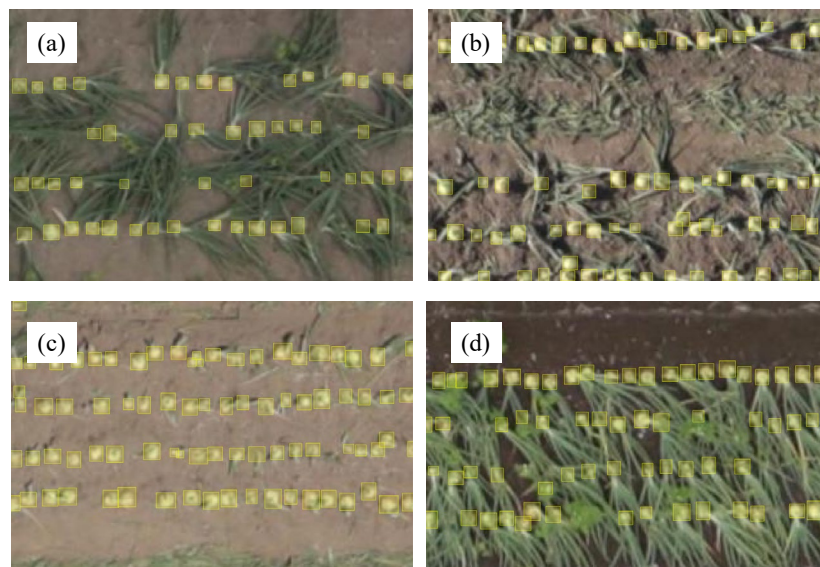
Figure 3. Examples of onion bulb annotation: (a) June 18th. (b) June 20th. (c) June 26th. (d) July 2nd.

3. RESULTS AND DISCUSSION

3.1 Orthomosaic

The series of orthomosaic and DEM are shown in figure 4 and figure 5. Obviously the DEM of June 20th, 26th and July 2nd have sphere shape error. This should be improved if the number of GCP increases while the manual process should be tedious. This is coursed by the wrong EXIF information of the camera. The camera was set just below the drone, the GPS condition wasn't good. It was revealed that some waiting period is necessary for stable GNSS signal condition for the camera. The rest DEM show similar land shape.

(a) June 18th (b) June 19th (c) June 20th (g) June 18th (h) June 19th (i) June 20th

(d) June 21th (e) June 26th (f) July 2nd (j) June 21th (k) June 26th (l) July 2nd
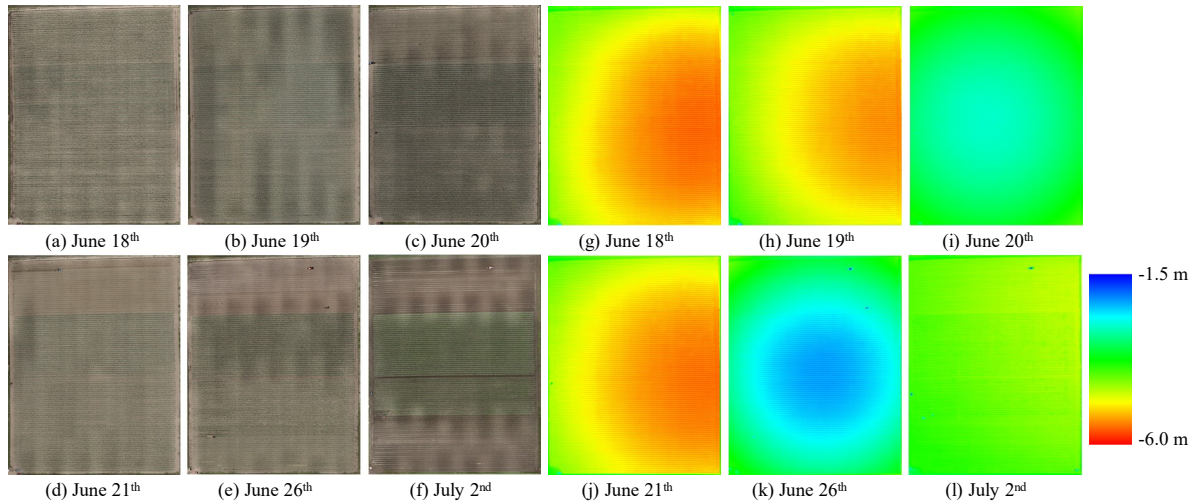
Figure 4 Orthomosaic and DEMs of onion field: (a) - (f) are orthomosaic images. (g) - (l) are DEMs.

Histograms of the altitude data of each pixel is shown in Figure 5. Deference between group of 18th, 19th and 20th and the rest is observed clearly. From above, if the GPS time in EXIF is correct, it is possible to obtain precise orthomosaic and DEM with only three GCPs.
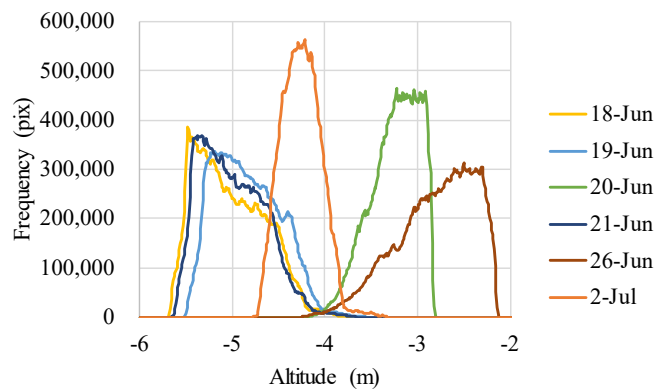
Figure 5 Histograms for each altitude data of DEM.

3.2 Onion bulb counting

The training image weren't sampled if the image didn't include onion bulb. As a result, number of false positive increased on grass area of neighboring the field. After detection of onion bulbs using deep learning, detected onion regions which is out of the field was eliminated. On June 26th and July 2nd, part of the field was empty after harvesting operation. The number and the estimated detection rate is shown in table 1. the number of the onion bulb ranged from 29,970 to 109,694 which were from 5 % to 19 % of the estimated onion plant number. As shown in figure 6, halves of bulbs weren't detected. It was revealed that the false-negative value in the confusion matrix should be reduced.

Table 1 Result of onion bulb counting.

Date	June 18	June 19	June 20	June 21	June 26	July 2
Onion bulb count	64,492	29,970	109,694	47,748	57,629	45,107
Row count	107	107	107	107	80	51
Plant count	564,960	564,960	564,960	564,960	422,400	269,280
Detection rate (%)	11	5	19	8	14	17
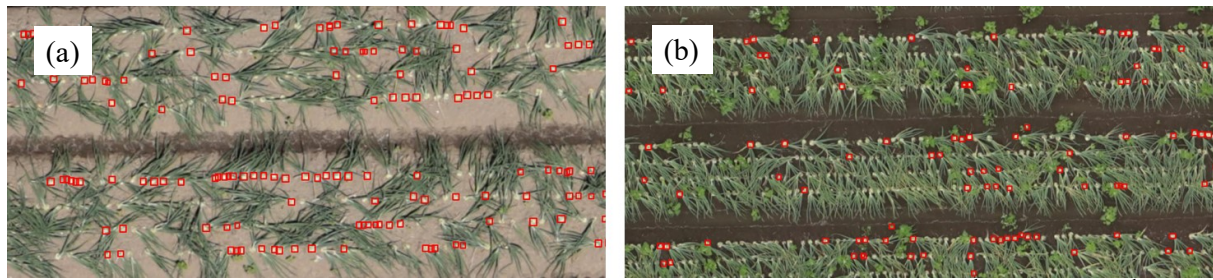
Figure 6 Examples of detecting onion bulbs using deep learning: (a) June 18th . (b) July 2nd .

4. CONCLUSION

We captured about 400 images of 6k resolution in a large-scale onion field using drone with RTK-GNSS. When the image localization was conducted successfully, precise orthomosaic and DEM were obtained with minimum number of GCPs. We then counted the number of onion bulbs applying deep learning for detection. The accuracy was low, however, our next step would be to improve the trained model by reducing the false-negative value in the confusion matrix.

ACKNOWLEDGMENT

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Oral Session | Food Safety

[5-1015-A] Food Safety (2)

Chair:Ubonrat Siripatrawan(Chulalongkorn University, Thailand)
Thu. Sep 5, 2019 10:15 AM - 11:30 AM Hall A (Main Hall)

[5-1015-A-04] Cinnamon Oil Nanoemulsion as a Natural Microbial Decontaminant of Chilled Fish Flesh

Piyanan Chuesiang[1,2], Romanee Sanguandeekul[1], *Ubonrat Siripatrawan[1,2]　（1. Chulalongkorn University, Department of Food Technology, Faculty of Science(Thailand), 2. The Novel Technology for Food Packaging &Control of Shelf Life Research Group, Chulalongkorn University(Thailand)）

10:15 AM - 10:30 AM

[5-1015-A-02] Application of Fluorescence Spectroscopy for the Classification of honey based on Geographical Origin

*Abdullah Iqbal[1,2], Mizuki Tsuta[1]　（1. Food Research Institute, National Agriculture and Food Research Organization 2-1-12 Kan-nondai, Tsukuba, Ibaraki 305-8642 Japan (Japan), 2. Dept. of Food Technology &Rural Industries, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh(Bangladesh)）

10:30 AM - 10:45 AM

[5-1015-A-03] Preservation of sardine and scallop by high hydrostatic pressure: safety and quality aspects

*Amauri Rosenthal[1], Rosiane Costa Bonfim[1,2], Fabiano Alves Oliveira[3], Ronoel Luiz de Oliveira Godoy[1], Carlos Adam Conte Junior[4], Eduardo Henrique Miranda Walter[1]　（1. Embrapa(Brazil), 2. Federal Rural University of Rio de Janeiro(Brazil), 3. Cefet Valença(Brazil), 4. Federal Fluminense University(Brazil)）

10:45 AM - 11:00 AM

[5-1015-A-01] Assessment of the Handling and Temporary Storage of Yams in Market Places in Ibadan, Oyo State, Nigeria

*Okwunna Maryjane Umego[1], Habeeb Adedotun Alabi[2], Yahaya Mijinyawa[2]　（1. Federal University Oye Ekiti(Nigeria), 2. University of Ibadan(Nigeria)）

11:00 AM - 11:15 AM

[5-1015-A-05] Responsiveness to Food Safety Emergencies in Eswatini following the Outbreak of listeriosis in South Africa

*Tendekayi Henry Gadaga[1], Anthony N Mutukumira[2]　（1. University of Eswatini(Swaziland), 2. Massey University(New Zealand)）

11:15 AM - 11:30 AM

10:15 AM - 10:30 AM (Thu. Sep 5, 2019 10:15 AM - 11:30 AM Hall A)

[5-1015-A-04] Cinnamon Oil Nanoemulsion as a Natural Microbial Decontaminant of Chilled Fish Flesh

Piyanan Chuesiang[1,2], Romanee Sanguandeekul[1], *Ubonrat Siripatrawan[1,2] （1. Chulalongkorn University, Department of Food Technology, Faculty of Science(Thailand), 2. The Novel Technology for Food Packaging &Control of Shelf Life Research Group, Chulalongkorn University(Thailand)）
Keywords: Essential oil, Nanoemulsion , Phase inversion temperature, Antimicrobial , Cell morphology

Economic losses caused by foodborne pathogen and spoilage are a driving force to apply food preservatives in perishable food products. However, the increasing awareness in recent years of the health risks for chemical preservatives added to the increasing demands of consumers for natural antimicrobial agents. This study aimed to develop cinnamon (*Cinnamomum verum*) essential oil nanoemulsion (CEO-NE) as a natural fledgling microbial decontaminant of a chilled fish product. The optimum CEO-NE formulation contained cinnamon essential oil with medium chain triglyceride (MCT) = 10 wt%, a non-ionic surfactant (Tween 80) =15 wt%, and deionized water 75 wt%. The CEO-NE was fabricated using a low energy Phase Inversion Temperature (PIT) method. Sea bass fish flesh was used to represent a seafood product. The fish flesh was artificially contaminated with *Escherichia coli* (ATCC 25922) prior to dipping into the CEO-NE solution at its minimum inhibitory concentration (MIC) determined from the previous experiments. The samples were stored at 4 C. The growth of *E. coli* and total viable counts of the CEO-NE treated samples was examined in comparison to those treated with bulk CEO and untreated (control) samples. The results showed that CEO-NE effectively inhibited *E. coli* and total aerobic bacteria better than bulk CEO. The bacterial cell morphological deformation by the CEO-NE was evidenced by field emission scanning electron microscopy (FE-SEM). The antimicrobial activity of the CEO-NE against *E. coli* was attributed to its ability to disrupt bacterial cell wall structures and promote expulsion of internal cellular material. The results suggest that the encapsulation of cinnamon oil in nanoemulsion enhanced its bactericidal activity against the targeted foodborne microorganism. The developed CEO-NE has potential to be used as natural antimicrobial agent for ensuring food safety of fish flesh or other seafood products.

Oral Session | Food Safety
[5-1015-A] Food Safety (2)
Thu. Sep 5, 2019 10:15 AM - 11:30 AM Hall A (Main Hall)

[5-1015-A-02] Application of Fluorescence Spectroscopy for the Classification of honey based on Geographical Origin

*Abdullah Iqbal[1,2], Mizuki Tsuta[1]　（1. Food Research Institute, National Agriculture and Food Research Organization 2-1-12 Kan-nondai, Tsukuba, Ibaraki 305-8642 Japan (Japan), 2. Dept. of Food Technology &Rural Industries, Bangladesh Agricultural University, Mymensingh-2202, Bangladesh(Bangladesh)）
Keywords: Honey, chemometrics, classification, geographic origin

The Front-face fluorescence spectroscopy was applied in this study for the classification of honey based on geographical origin. Honey samples (*Robinia pseudoacacia* and Blended floral source) of different origin (i.e., China, Hungary and Japan etc) used in this study were collected from their production sites. Before the fluorescence measurement, the samples were put in shaking water bath at 60℃ for 30 min with 100 rpm shaking speed. Then after stirring to obtain the homogeneity, the honey samples were diluted to 100 times with the addition of 20% (v/v) ethanol solution. The front-face fluorescence excitation-emission matrices were then recorded from 200nm to 800nm (at an interval of 1 nm) whereas excitation spectra were recorded between 200nm to 500nm (with an interval of 5nm). With the application of necessary pre-processing (i.e., normalization, mean centering, autoscaling and/or combination thereof) and digital smoothing polynomial filters (i.e., Savitzky-Golay smoothing filters) for smoothing out the noisy signals, the rayleigh scattering rays were removed from the spectra. The chemometric analysis were then applied to the spectral data using principal component analysis (PCA) and partial least squares-discriminant analysis (PLS-DA) for classification of the honey samples. A reasonable sensitivity (ranging from 0.90 to 1.000) and specificity (ranging from 0.795 to 1.000) for class predictions was obtained from the PLS-DA model. The results showed that front-face fluorescence spectroscopy has potential for the discrimination of *Robinia pseudoacacia* honey based on geographical origin. But it is not possible to discriminate the blended samples based on geographical origin.

Application of Fluorescence Spectroscopy for the Classification of Honey Based on Geographical Origin

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ABSTRACT

The Front-face fluorescence spectroscopy was applied in this study for the classification of honey based on geographical origin. Honey samples (*Robinia pseudoacacia* and Blended floral source) of different origin (i.e., China, Hungary and Japan etc) used in this study were collected from their production sites. Before the fluorescence measurement, the samples were put in shaking water bath at $60°C$ for 30 min with 100 rpm shaking speed. Then after stirring to obtain the homogeneity, the honey samples were diluted to 100 times with the addition of 20% (v/v) ethanol solution. The front-face fluorescence excitation-emission matrices were then recorded from 200nm to 800nm (at an interval of 1 nm) whereas excitation spectra were recorded between 200nm to 500nm (with an interval of 5nm). With the application of necessary pre-processing (i.e., normalization, mean centering, autoscaling and/or combination thereof) and digital smoothing polynomial filters (i.e., Savitzky-Golay smoothing filters) for smoothing out the noisy signals, the rayleigh scattering rays were removed from the spectra. The chemometric analysis were then applied to the spectral data using principal component analysis (PCA) and partial least squares-discriminant analysis (PLS-DA) for classification of the honey samples. A reasonable sensitivity (ranging from 0.90 to 1.000) and specificity (ranging from 0.795 to 1.000) for class predictions was obtained from the PLS-DA model. The results showed that front-face fluorescence spectroscopy has potential for the discrimination of *Robinia pseudoacacia* honey based on geographical origin. But it is not possible to discriminate the blended samples based on geographical origin.

Keywords:
Honey, chemometrics, classification, geographic origin, PCA, PLS-DA

1. INTRODUCTION

Honey is a healthy natural, pure and nutritious food produced by honeybee containing 60–80% of carbohydrates, 17–20% of water, 0.3–0.8% of proteins, 0.2% of minerals and minor quantities of amino acids, phenols, pigments, vitamins, volatile substances, and others (Ball, 2007, Bogdanov et al., 2008, Khan et al., 2017).

Traditionally, honey has been used by human being not only as a nutritious substance but also as a therapeutic product due to its antioxidative components, such as polyphenols, amino and organic acids, enzymes and proteins (Oryan et al., 2016). These components are highly dependent on the floral source, the geographical region of production and external factors associated with environmental conditions, processing and storage methods (Alzahrani et al., 2012). Regional and/or geographical characteristics of honey in terms of the composition varies depending on the climate, altitude and other environmental factors etc (Salonen et al., 2017). Therefore, the geographical origin play an important role in the overall quality and authenticity of honey which is essential to be considered for quality point of view.

Recently, authenticity of foodstuffs became a major issue for the consumers and producers worldwide (Petróczi et al., 2010). In case of honey, authenticity is related to both geographical and floral source determinations as well as detection of unwanted substances, like syrups or sugars. Geographical

origins are economically important and therefore, subjected to frauds, leading to false or doubtful labelling. During the last two decades, several researchers attempted to characterize the botanical and geographical origin of honey by exploiting different analytical techniques (Anklam, 1998), such as FTIR (Wang et al., 2010), FT-Raman spectroscopy (Corvucci et al., 2015), mid-infrared spectroscopy (Ruoff et al., 2006), near-infrared spectroscopy (Woodcock et al., 2007), and fluorescence spectroscopy (Lenhardt et al., 2015, Mehretie et al., 2018). In many cases, several analytical methods are simultaneously essential for a reliable authentication of geographical origin of honeys which is time-consuming, laborious, costly and requires vast technical skill. Therefore, there is a need for new methods which can provides a rapid and reproducible authentication of the geographical origin of honey. As a result, the determination of botanical and geographical origin of honey is of increasing interest worldwide. The use of excitation-emission matrix (EEM) seems to be a promising approach as it has been successfully applied for different products. Fluorescence spectroscopy provides information on the fluorescent molecules' presence and the environment of honey produced like other biological samples. Hence, fluorescence spectroscopy seems to be effective for classification of honey based on geographical origin. Therefore, the aim of the present research is to classify honey collected from different geographical origins using front-face fluorescence spectroscopy.

2. MATERIALS AND METHODS
2.1 Sample Collection and Preparation for Measurement
A total of 23 honey samples (*Robinia pseudoacacia*) and 49 samples (Blended) produced in different countries (i.e., China, Hungary and Japan, Canada, Argentina and Myanmar) were used in this study. The honey samples were collected from their production sites and stored at 4°C until analysis. Before the fluorescence measurement, the samples were put in shaking water bath at 60℃ (for liquefaction) for 30 min with 100 rpm shaking speed. Then after stirring to obtain the homogeneity, the honey samples were diluted to 100 times with the addition of 20% (v/v) ethanol solution. Then 3ml of diluted samples were pipetted into quartz cuvette and placed in the sample holder. The excitation-emission matrices were then recorded with the fluorescence spectrometer (F–7000, Hitachi High–Technologies Corporation, Tokyo, Japan), from 200nm to 800nm (at an interval of 1 nm), whereas excitation spectra were recorded between 200nm to 500nm (with an interval of 5nm). The spectra were then converted into ASCII files to be further analyzed using MATLAB®2019a (The MathWorks, Inc., Natick, MA).

2.2 Processing of Spectra and Multivariate Analysis
With the application of necessary pre-processing (i.e., normalization, mean centering, autoscaling and/or combination thereof) and digital smoothing polynomial filters (i.e., Savitzky-Golay smoothing filters) for smoothing out the noisy signals, the Rayleigh scattering rays were removed from the spectra using a script coded and designed on MATLAB®2019a (The MathWorks, Inc., Natick, MA). The chemometric analysis such as Principal component analysis (PCA) was used to eliminate the spectral collinearity, random noise, and to reduce the dimensionality of variables. It was also applied in visualizing the data set as well as exploring the variations among the sample classes. The data sets in PCA are not correlated with each other and the score plot of the PCA has been used to explain the variations or similarities among the samples (Rahman et al., 2016). Subsequently, the partial least squares-discriminant analysis (PLS-DA) was used for classification of the honey samples. PLS-DA is a classification technique used for building linear discriminant analysis transforming the observed data into a set of intermediate linear latent variables which are then used for predicting the dependent variables. To select the number of PLS variables included in the model, Venetian blind cross-validation was used in this investigation. PCA and PLS-DA of the samples were computed by using the PLS toolbox for MATLAB (Eigenvector Research Inc., Wenatchee, WA, USA).

3. RESULTS AND DISCUSSION
3.1 Spectral information
The excitation-emission matrix (EEM) for typical geographical samples after removing Rayleigh's scattering rays from the spectra are shown in Figure 1(a). It is seen that all the honey samples irrespective of geographical origin, shows three peaks in the contour plot of EEM. The peak exists at around excitation of 230 nm and emission of 340 may be responsible for the fluorescence of aromatic

amino acids (Karoui et al., 2007) present in the honey. The excitation/emission wavelengths corresponding to the peak 280/340 nm, common to all three samples may be due to the presence of flavonoids (such as apigenin, chrysin, kaempferol, pinocembrin), although it is tough to identify the flavonoids responsible for such peaks (Lenhardt et al., 2015). Another fluorescence peak corresponding to excitation wavelength range of 320-340 nm and emission wavelength range of 400-460 nm could be related to the Maillard reaction products such as hydoxymethylfurfural and furosine (Lenhardt et al., 2015). The contour plot of spectra for the *Robinia pseudoacacia* samples generally indicates that the considered honey samples may have similar components although they are originated or produced in different countries. However, the variation in peaks may be due to the concentration of different components.

The spectral behavior of blended honey samples is bit complex and there are distinct differences among the samples. Even the samples blended in the same geographical location (i.e., samples from same country), the different blended samples gave different fluorescence signatures as shown in figure 1(b). This may be due to the ingredient of the blended components and their concentration as well as other parameters associated with the blending process which cannot be explained unknown during the investigation.
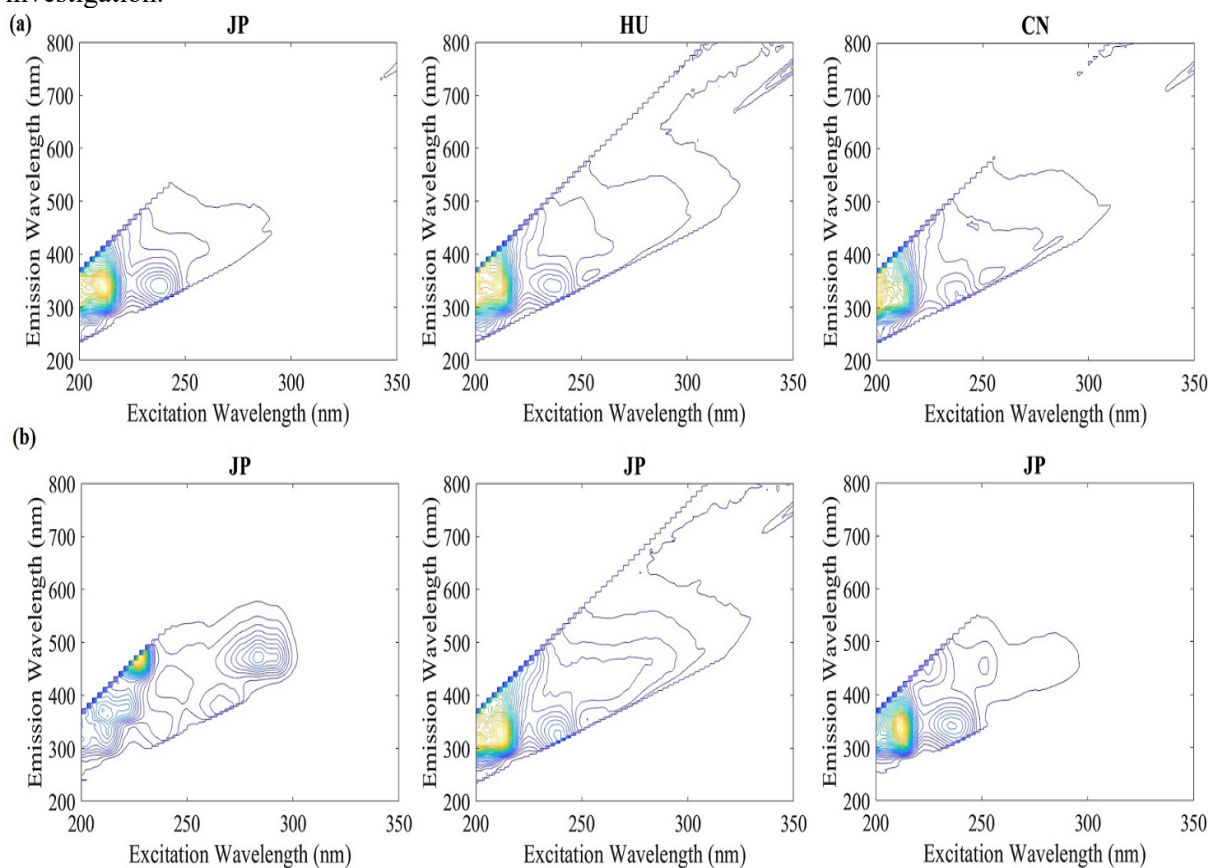
Figure 1. Excitation-emission spectra of different honey[*]: (a) Robinia pseudoacacia, (b) Blended
([*]JP=sample from Japan, HU= sample from Hungary, CN=sample from China)

3.2. Chemometric analysis
3.2.1 Principal component analysis (PCA)
PCA is applied for both types of honey samples (*Robinia pseudoacacia* and blended) to reduce multidimensionality to two dimensions and the results are shown in Figure 2. From the PC scores it is seen that for the pseudoacacia samples (Figure 2.a), the PC1 explained 63.80% of the total variance in the data set while PC2 explained 17.46% and remaining 18.74% of data variance belongs to the other dimensionality. From the score plot, the honey samples are not completely separated into different classes or groups. It is seen from the plot that they are very close to each other (as it is mentioned earlier that showing similar peaks). The similar but more prominent behavior has been observed for

the blended sample as shown in Figure 2(b), although the PC1 explained 61.23% of the total variance in the data set while PC2 explained 18.33% whereas remaining 20.44% of data variance belongs to the other PCs.
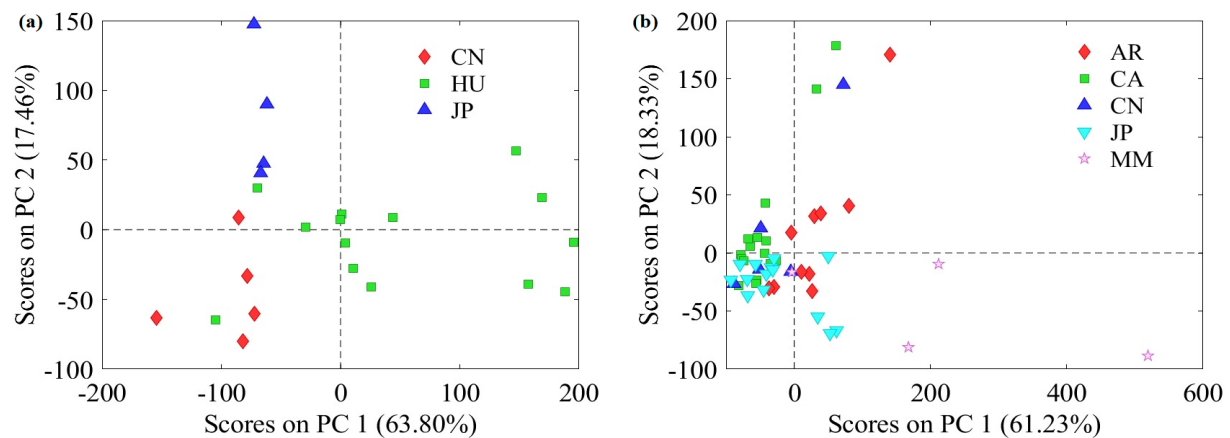
Figure 2. Two dimensional PCA score plots of honey[*]: (a) Robinia pseudoacacia (b) Blended
([*]CN=sample from China, HU=sample from Hungary, JP= sample from Japan, CA=sample from Canada, MM=sample from Myanmar)

3.2.2 Partial least squares-discriminant analysis (PLS-DA)

The PLS-DA classification model was developed to classify different honey samples based on geographical origin and four parameters such as sensitivity, specificity for calibration (Cal), and cross-validation (CV) were considered as the indicator of the robustness of the model (Table 1).

Table 1. Sensitivity, specificity, and classification error of PLS DA models.

Parameters*	Robinia pseudoacacia	Blended
Sensitivity (Cal)	1.000	0.900
Sensitivity (CV)	1.000	0.900
Specificity (Cal)	1.000	0.872
Specificity (CV)	0.944	0.795

*Cal refers to calibration set, and CV refers to the cross-validation results

The best model was built with 5 and 4 latent variables for pseudoacacia and blended samples, respectively. The models showed reasonable sensitivity (from 0.900 to 1.00 for both Cal and CV) and specificity (0.795 to 1.00 for Cal and 0944 for CV) for the classification of honey samples for both the the Pseudoacacia and blended samples (Table-1). The confusion table (CV) for both types of honey samples are shown in Table 2.

Table 2. Confusion table for honey samples

Predicted as...	Actual Class (Robinia pseudoacacia)				Predicted as…	Actual Class (Blended)				
	CN (5)	HU (14)	JP (4)			AR (10)	CA (16)	CN (5)	JP (14)	MM (4)
China (CN)	5	1	0		Argentina (AR)	9	3	1	2	0
Hungary (HU)	0	13	0		Canada (CA)	1	8	2	2	0
Japan (JP)	0	0	4		China (CN)	0	4	1	0	0
					Japan (JP)	0	1	0	10	0
					Myanmar (MM)	0	0	1	0	4

From table-2, it is seen that *Robinia pseudoacacia* samples can be classified based on geographical origin. But for the blended honey, samples only from Argentina and Myanmar, has been classified as the origin they belong. Remaining samples are mixed with other classes which seems to be like that group(s) as described in section 3.1.

4. CONCLUSION

This study was conducted to demonstrate the potential application of excitation-emission matrix (EEM) patterns for the classification of complex matrix of honey samples from different geographical origin. The front face fluorescence measurement used in this study revealed that it is not possible to obtain complete classification of honey based on geographic origin by Front-face fluorescence spectroscopy. Although the classification accuracy for pseudoacacia samples was reasonable but not up to the level for the blended samples to conclude with a sentence 'complete classification' is possible! However, further studies need to be carried out with the modification of existing Front-face fluorescence spectroscopy with more samples to make a conclusion about the potential for the classification of honey based on geographic origin.

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10:45 AM - 11:00 AM (Thu. Sep 5, 2019 10:15 AM - 11:30 AM Hall A)

[5-1015-A-03] Preservation of sardine and scallop by high hydrostatic pressure: safety and quality aspects

*Amauri Rosenthal[1], Rosiane Costa Bonfim[1,2], Fabiano Alves Oliveira[3], Ronoel Luiz de Oliveira Godoy[1], Carlos Adam Conte Junior[4], Eduardo Henrique Miranda Walter[1] (1. Embrapa(Brazil), 2. Federal Rural University of Rio de Janeiro(Brazil), 3. Cefet Valença(Brazil), 4. Federal Fluminense University(Brazil))
Keywords: Sardine, Scallop, High Hydrostatic Pressure, Preservation, Shelf life

High hydrostatic pressure (HHP) has been a successful novel technology for preservation of different foods, including seafood and fishes. However, safety and quality aspects have to be considered for designing a proper process aiming at optimizing the quality and assuring the safety of the products. Some studies have been carried out for comparing quality and safety aspects of sardine and "Lion Paw" scallop muscle processed by HHP. Therefore, sardine fillets and scallops muscle were treated by 300 MPa to 400 MPa for 0 to 15 min. and compared regarding microbiology, TBARS, N-TVB formation and nucleotide degradation along refrigerated (4-5°C) shelf-life. In the case of sardines, HHP did not completely cease N-TVB formation and nucleotide degradation, but minimized the development of those processes, especially at higher pressure levels and holding times. Regarding scallops, HHP decreased the count of mesophilic and psychotropic microorganisms below the legislation standard requirements. However, proper caution should be taken mainly considering specific pathogenic microorganisms. As expected, HHP accelerated lipid oxidation in the case of scallops, resulting in increase of TBARS, but did not exceed the standard limit of 2 mg/kg. Nucleotide degradation followed different patterns considering the different metabolisms and specificities of the muscle fibers. These results indicate that HHP can significantly increase the refrigerated storage time for sardine and scallop but intrinsic and extrinsic factors and characteristics may influence the safety and quality aspects.

Oral Session | Food Safety
[5-1015-A] Food Safety (2)
Thu. Sep 5, 2019 10:15 AM - 11:30 AM Hall A (Main Hall)

[5-1015-A-01] Assessment of the Handling and Temporary Storage of Yams in Market Places in Ibadan, Oyo State, Nigeria

*Okwunna Maryjane Umego[1], Habeeb Adedotun Alabi[2], Yahaya Mijinyawa[2] (1. Federal University Oye Ekiti(Nigeria), 2. University of Ibadan(Nigeria))
Keywords: Yam, Yam Storage, Open Shed, Yam Stall

Yams are scarce during non-harvest seasons and the prices are exorbitant with majority of the population unable to buy. This situation motivated the interest for this research to assess the handling of yam and the temporal storage practices among traders in order to identify and have good understanding of the various activities pertaining to the yam markets. Visits to the markets, interview with the traders and measurement of the storage temperature and relative humidity were carried out to obtain data for the assessment of handling and temporal storage of yams in the markets. Five activities were identified pertaining to yams in the markets, namely: arrival of yams in vehicles, unloading of the yams, display of the yams for sale, packaging and loading of sold yams, and lastly temporal storage of the unsold tubers. The assessment of the handling of yam tubers in each of the above mentioned activities revealed that; the handling operations are rudimentary and results in bruising, breakage and exposure of tubers to adverse environmental conditions thereby causing substantial losses. The assessment of the temporal storage structures for yams in the markets showed that; there are two types of storage structures for yams in the markets, these are: the open shed and the market stalls. The storage environment, the design and construction materials of these storage structures are not effective for yam, thereby contributing to losses. These findings revealed that the open shed and market stall rooms used by yam wholesalers in Bodija and Bere-Mapo markets are ineffective for yam storage because the storage environment within these structures as influenced by the design and construction materials cannot allow for effective storage of yams. The problems associated with these structures in percentage are roof leakage 34.8% and 11.4%, rodent and pest attacks 82.6% and 11.4%, and adverse environmental conditions 91.3% and 85.7% for open sheds and stalls respectively. It is recommended that the materials of construction and design of these structures be modified to make them more effective.

Assessment of the Handling and Temporary Storage Methods of Yams in Market Places in Ibadan

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ABSTRACT

Yams are scarce during non-harvest seasons and the prices are exorbitant with majority of the population unable to buy. This situation motivated the interest for this research to assess the handling of yam and the temporal storage practices among traders in order to identify and have good understanding of the various activities pertaining to the yam markets. Visits to the markets, interview with the traders and measurement of the storage temperature and relative humidity was carried out to obtain data for the assessment of handling and temporal storage of yams in the markets. Five activities were identified pertaining to yams in the markets, namely: arrival of yams in vehicles, unloading of the yams, display of the yams for sale, packaging and loading of sold yams, and lastly temporal storage of the unsold tubers. The assessment of the handling of yam tubers in each of the above mentioned activities revealed that; the handling operations are rudimentary and results in bruising, breakage and exposure of tubers to adverse environmental conditions thereby causing substantial losses. The assessment of the temporal storage structures for yams in the markets showed that; there are two types of storage structures for yams in the markets, these are: the open shed and the market stalls. The storage environment, the design and construction materials of these storage structures are not effective for yam, thereby contributing to losses. These findings revealed that the open shed and market stall rooms used by yam wholesalers in Bodija and Bere-Mapo markets are ineffective for yam storage because the storage environment within these structures as influenced by the design and construction materials cannot allow for effective storage of yams. The problems associated with these structures in percentage are roof leakage 34.8% and 11.4%, rodent and pest attacks 82.6% and 11.4%, and adverse environmental

conditions 91.3% and 85.7% for open sheds and stalls respectively. It is recommended that the materials of construction and design of these structures be modified to make them more effective.

1. INTRODUCTION

Nigeria is the largest producer of yams in the world, annually producing about 31 million tonnes. Nigeria produced 60% of the world's yams in 2010, and is the largest contributor in Africa's "Yam Belt," a yam production area that comprises Nigeria, Ghana, Benin, Côte d'Ivoire, Central African Republic, Cameroon, and Togo. Yams have had the second highest production level of any food crop in Nigeria in the past 50 years after cassava (Bergh *et al.*, 2012). Yam losses are one of the greatest problems facing yam production in Nigeria and are of concern to everyone, from the research scientists to the extension workers, marketers in the field to the farmers on the farm and to the government policy formulators. The post-harvest handling and storage practices for yams in Nigeria presents a dismal picture and are mostly comprised of traditional techniques practiced by growers, traders and the processor resulting in considerable deterioration of physical and nutritional qualities of harvested crop (Oni and Obiakor, 2002). Interest in the reduction of post-harvest losses is not new. Mrema and Rolle (2002) reported that after the mid-1970s food crisis, the United Nations brought post-harvest storage losses into international focus in 1975 when it declared that "further reduction of post-harvest food losses in developing countries should be undertaken as a matter of priority". In underdeveloped and developed tropical countries, both quantitative and qualitative losses of agricultural products occur at all stages in the post-harvest chain, from harvesting, through handling, storage, processing, packaging, transportation and marketing until crops are delivered to the final consumers.

Ibadan North local Government is a big urban center with a population of over 350,000 inhabitants, according to the 2006 Nigerian census (NPC, 2006). The town is home to two major urban yam markets in Oyo State, that is, Bodija and Bere-Mapo yam markets. According to the Natural Resources Institute (NRI, 2012) report, diverse challenges constrain yam farmers and marketers' ability to fully exploit the potential of yams and yam products in the southwest, these includes, high cost of inputs and labour, lack of credit, limited access to proper secure storage facilities, high transportation costs and ineffective handling practices. The yam traders in Ibadan North are no exception to the above mentioned challenges.

Despite the elaborated agricultural programs, Ibadan is still unable to provide an all year round supply of yams within the purchasing power of majority of the people. Besides economic factors, the supply of food in the local government is limited by losses due to wastage and spoilage. Though no one knows how much yams is lost between harvest and consumption, but post-harvest management complements efforts to enhance food security through improved farm level productivity, thus tending to benefit producers, and more specifically, the rural farmers.

Post-harvest management reduces post-harvest losses thus, generates income, improves product quality and safety, and contributes to food and nutritional security. It is against this background that an analysis of the post-harvest management strategies like handling and temporary storage by yam traders is deemed important. This work assessed the handling and temporary storage of yams in markets in Ibadan North local government

2. METHODOLOGY

2.1 Study Area

This study was carried out in Ibadan North Local Government area. The city of Ibadan is located approximately on longitude 3^05" E of the Greenwich Meridian and latitude 7^023" N of the Equator. Economic activities undertaken by people in the Local Government Area include trading, public service and agriculture. Ibadan North Local Government has a land area of $145.58km^2$ and a population of 306,795 people (NPC, 2006). The study area experiences a tropical type of climate. It has a mean annual temperature of about 32^0 C. The relative humidity can be as high as 95% and a total of about 1250 mm as mean annual rainfall.

2.2 Methodology

The markets were visited for physical observation of the activities taking place, particularly among the yam wholesalers. Also, the yam storage structures were assessed. Temperature and relative humidity of the storage structures and the ambient environment were measured once every other day for a period of one month (August 19th to September 16th, 2016). A dry bulb and wet bulb thermometer with psychrometric chart was used to achieve this. A questionnaire was designed to obtain information on some of the questions regarding yam storage among yam wholesalers in Bodija and Bere-Mapo markets. The data collected was analyzed using the Microsoft Excel 2010 to obtain statistics of frequencies and percentages of the data.

3. RESULTS AND DISCUSSIONS

3.1 Arrival of Yams to the Markets

Yams are transported from the farms or small local district markets in rural areas to the large urban markets of Bodija and Bere-Mapo in big lorries, buses, open pick-up vans, and trucks. It was observed that, yams are stacked one upon another like timber, without any packaging material. Yams that are in contact with the edges of the vehicle sustained abrasions and cuts, those at the bottom are subjected to compressive loads due to the weight of the overburdening yams lead to internal injury or damage of the yams at the bottom. Depending on the degree of injury on the yam, the level of periderm formation might be affected. When the periderm is not formed the yam cannot heal the bruised part, thus, the storage life is reduced.

3.2 Unloading Operation

This operation is carried out manually by the market labourers. The labourers unload the vehicles either by using metal pans to pack the yams from the vehicle or by throwing the yams from the vehicle to other labourers standing on the ground who then place it on the ground gently. Unloading by throwing if not done carefully can lead to breakage (figure 1). In the case of unloading with the metal pan, there is risk of compression damage due to the force acting on individual tubers. It was also observed that the labourers carry plenty yams at a time and get fatigue under the weight. Thus, instead of gently putting down the tubers they drop it by pouring. This results in tuber bruises and breakages.

Figure 1: Unloading operation by throwig

3.4 Packaging and Loading of Sold Yams

Sometimes the market labourers are contracted to carry the yams to the vehicle. The practice of loading yams in the vehicle by pouring before arranging as shown in figure 2 is damaging to the yams. Some yams break and others get bruised and as a result such tubers do not take long to spoil. As recommended by Ayoub and Lennox (2013), packaging materials such as telescopic fiberboard cartons with paper wrapping or excelsior should be used. This reduces bruising and damage due to heat from the tuber respiration and breakage and internal injury caused by compression of tubers from the weight of the overlying tubers.

Figure 2: Loading operation by pouring

3.5   Temporal Storage of Yams in Markets

The markets are not used for long storage of yams, however, there is yam storage in the markets on a temporary basis. This is because tubers are usually supplied in very large quantities and the supplies are usually not exhausted in a few days. Sometimes it takes over a month before some traders are able to exhaust their supplies. Thus, there is storage of the produce while the stock lasts.

3.5.1   Types of Yam Storage Structures

There are only two types of yam storage structures in Bodija and Bere-Mapo markets in Ibadan North local government, Oyo State. In Bodija yam market, wholesalers use open sheds (figure 3) to store yams, while those in Bere-Mapo yam market, use stalls as structures for yam storage (figure 4). These structures can best be described as improvised yam storage structures as the design and types of construction materials are not in tandem with any known design criteria or principle for yam storage structures. However, traders have been using these structures for years for the storage of yams. These structures were assessed to see how they vary from known traditional and modern yam storage structures, and what improvements they need to become effective in storing yams.

Figure 3: Open shed

Figure 4: Yam Stall

3.5.2   Storage Environment of the Structures

The average daily ambient temperatures and the temperatures inside the open shed in Bodija and the market store rooms in Bere-Mapo yam markets are presented in figures 5 and 6. While the temperatures in the open shed storage structures varied from 25.3^0C to 30.3^0C with an average value of 28.03^0C and an average ambient temperature of 28.67^0C, those within the stall rooms varied from 30.1^0C to 33.8 ^{0}C with an average value of 33.21^0C, and for the ambient temperatures, the range is from 26.0^0C to 33.0^0C with an average value of 29^0C. The temperatures within the open shed was generally equal to that of the ambient conditions, but lower than those obtained in the market stall rooms for all periods throughout the study

period. These average temperature values are higher than the storage temperature of 13^0C to 15^0C for yam recommended by the National Agriculture Research Institute, 2004.

The ambient relative humidity ranged from 76% to 82% with an average value of 79.73%, while for the open shed, it varied from 79% to 83% with an average value of 80.10%, and for the stall rooms it varied from 82% to 88% with an average value of 85.15%. Although, there were a few overlaps, the ambient relative humidity was equal to those in the open shed structure but lower than those within the market store rooms (figures 7 and 8). However, these average relative humidity are lower than the recommended value of 90% to 95% by the National Agriculture Research Institute, 2004.

The variation in the environmental conditions within the market stall rooms and the open shed structure is attributed to the lack of ventilation in the store rooms, material of construction and the arrangement of the yams in the store rooms. This observation is due to the fact that within the stall room storage structures, the respiration of the tubers of yam increased the internal temperatures of the structures which is not the case under the open shed environment.
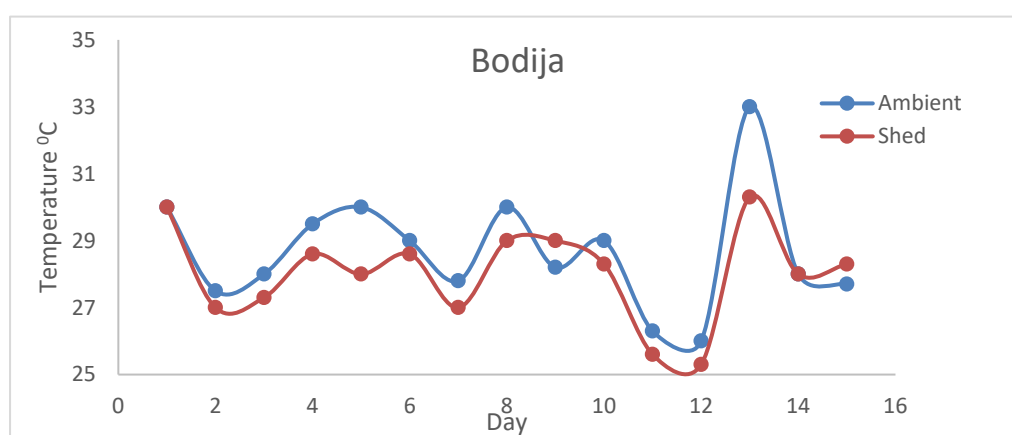
Figure 5: Average Daily Temperatures of Yam Storage Structures in Bodija
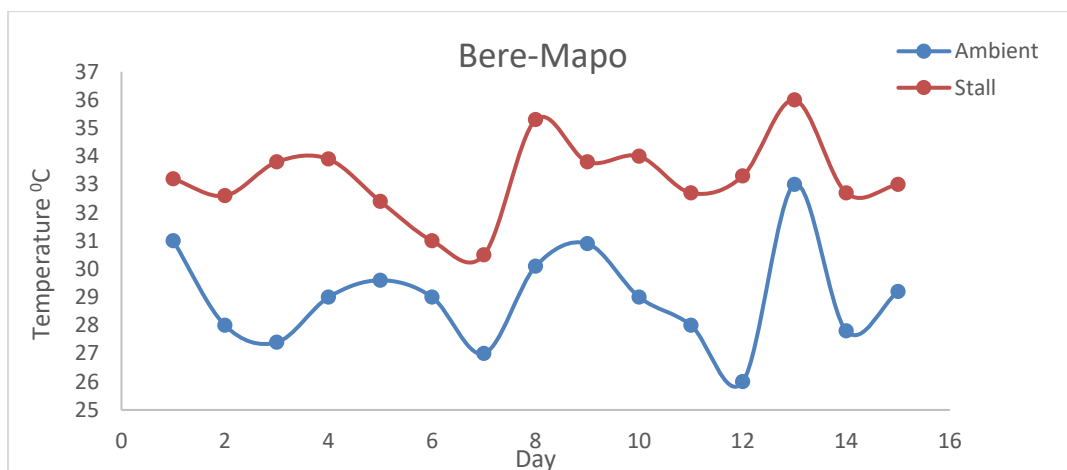
Figure 6: Average Daily Temperatures of Yam Storage Structures in Bodija
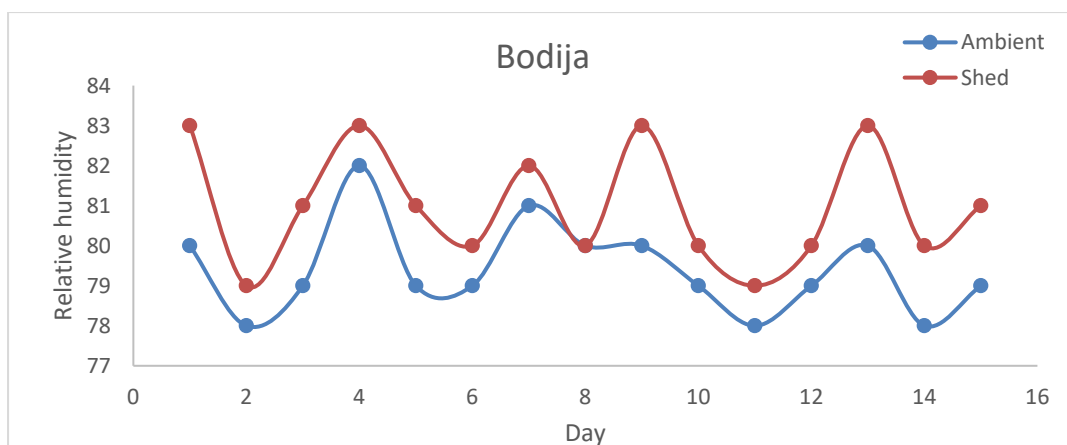
Figure 7: Average Daily Relative Humidity of Yam Storage Structures in Bodija
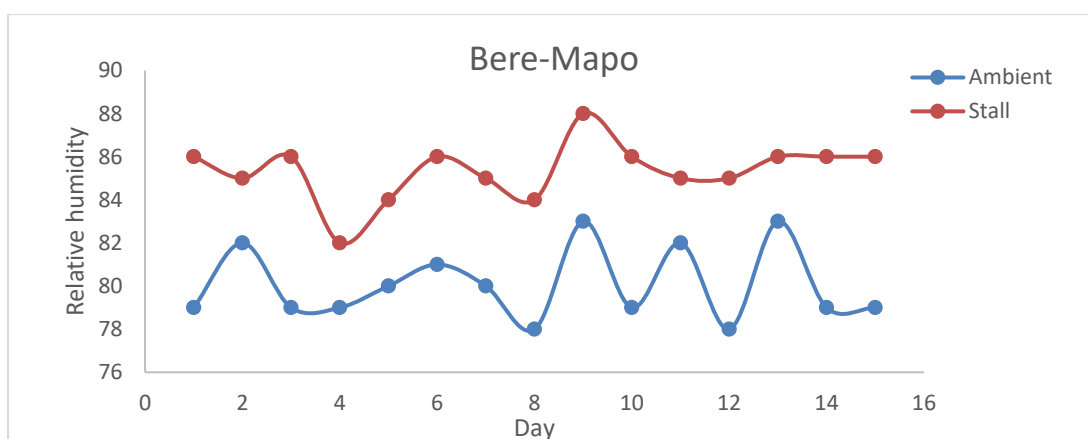
Figure 8: Average Daily Relative Humidity of Yam Storage Structures in Bere-Mapo

3.5.3　Storage Structure Construction Materials

The materials of construction are wood, sand and cement blocks walls, the roof is corrugated zinc or aluminum roofing sheets and ventilation is inadequate because only one opening, which is the door, is fitted on the store. These construction materials are not very good insulators of heat. The roofing sheets for example, easily conducts solar heat and transmits it easily into the room as no ceiling is provided and the height is low (2.3 meters). The arrangement of the yams in the stalls by heaping does not permit maximum air circulation between the tubers as compared to the arrangement in the open sheds. This is detrimental to the tubers because, during respiration of yams, oxygen is used and CO_2, water and heat are produced. Since there is no proper air circulation to transport the heat and water away from the tubers, the heat causes rise in temperature and water increase the moisture in the air, which is the relative humidity Physiological activities like respiration and sprouting of the tubers are promoted by high temperature of the storage environment which results in a steady loss of carbohydrate in the form of carbon dioxide and water, making the yams to lose weight, size and market value.

3.5.4　Problems Associated with the Temporal Storage Structures

The problems associated with yam storage structures in Bodija and Bere-Mapo yam Markets are presented in Table 1. The results indicate that environmental factors constitute a major problem in both markets. While for Bodija market, 91.3% of the respondents said storage environment conditions within the open shed was not favorable for yam storage, for Bere-Mapo market, 85.7% attributed yam storage losses to adverse environmental conditions within the store rooms. Decay was very high in tubers heaped on floor as a result of direct contact with the soil on the bare ground. Presence of rot pathogen in soil on the storage area serves as a source that initiates decay. Poor air circulation within the heaped yam aid in the build-up of heat and increase humidity as a result of respiration. Hence induces spore germination and growth of pathogens.

Another major problem is the incidence of pest and rodents attacks which is particularly high (82.6%) in the open shed, but low in the market stall rooms with 11.4%. This high variation is attributed to the fact that in the market stalls, the rooms are fumigated and the doors are closed and rat poisons are used in preventing rodents and other pests from damaging the stored yams. However, within the open sheds, fumigation is not effective because the structure has no enclosure. Rat poisons are used against rodent attacks, but soon afterwards,

another set of rodents migrate from the nearby refuse dump sites and bushes to attack the stored yams since they are kept in the open space.

Other storage problems identified are roof leakage and storage space. While in Bodija 34.8% of sheds assessed had the problem of roof leakage, those within Bere-Mapo is 11.4%. Roof leakage allows direct sun rays and rain water to impact on the stored tubers. The continuous heating and wetting of the tubers result in breaking yam dormancy period sooner than necessary. Once dormancy period is over, sprouting sets in. Sprouting of stored yams is not desired because it affects the nutrition and size of the tuber. It can also result to decay of the yam and after its viability. Also, roof leakage increases the relative humidity within the storage environment and in combination with high temperature, encourages mold growth and insect activity.

Table 1: Problems Associated with Yam Storage Structure in Bodija and Bere-Mapo Markets.

Problems	Frequency(n=23)	Bodija	Frequency(n=35)	Mapo
Leakages	8	34.8%	5	14.3%
Collapse	1	4.3%	-	-
Rodents Attack	19	82.6%	7	20%
Environmental Factors	21	91.3%	30	85.7%

4. CONCLUSION AND RECOMMENDATIONS

4.1 Conclusion

The assessment of the handling of yam tubers in each of the above mentioned activities reveals that; the handling operations are rudimentary and results in bruising, breakage and damage of the yams causing substantial losses.

Furthermore, this study reveals that, there are only two types of temporal storage structures for yams in Bodija and Bere-Mapo yam markets, that is, the open shed and market stalls structures respectively. The assessment of these methods of yam storage structures shows that they are not efficient methods for yam storage because, the design and types of construction materials used cannot significantly moderate the storage temperature and relative humidity of the stored yams. Also, the design does not provide the means for protecting the stored yam

tubers from rodents and other insects attack. Roof leakage, rodents attack, collapse, and harsh environmental conditions were some of the problems with these temporal storage structures

4.2 Recommendations

Proper handling of yams should be adopted both by farmers and wholesalers to minimize losses. The stall room used for yam storage should be well ventilated in order to enhance the exchange of air between the enclosure and the surroundings thereby eliminating the enzymatic action and micro-organism activities which result in the rapid spoilage of stored produce. Markets storage structures should be modified by adopting the design and types of construction materials recommended by Nigerian Stored Produce Research Institute (NISPRI).

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Oral Session | Food Safety
[5-1015-A] Food Safety (2)
Thu. Sep 5, 2019 10:15 AM - 11:30 AM Hall A (Main Hall)

[5-1015-A-05] Responsiveness to Food Safety Emergencies in Eswatini following the Outbreak of listeriosis in South Africa

*Tendekayi Henry Gadaga[1], Anthony N Mutukumira[2] (1. University of Eswatini(Swaziland), 2. Massey University(New Zealand))
Keywords: food safety, listeriosis, Eswatini, food control, pathogens

The FAO defines food safety as the absence, or safe, acceptable levels of hazards in food that may harm the health of consumers. Microbiological hazards pose a disproportionate threat to human health in all countries, more so in developing countries due to inadequate resources and fragmented food control systems. The food control system in Eswatini is administered by different departments. The Ministry of Health is responsible for the administration of the Public Helath Act; Ministry of Agriculture, the Dairy act, and the Ministry of Trade, Industry and Commerce, standards, including food standards. A 2013 Food Safety Bill that aims at coordinating food control activities under a single food control authority has yet to be finalised. The outbreak of listeriosis in South Africa in 2017 revealed the importance of an effective food control system in Eswatini. Under the current food control system, the country runs the risk food poisoning outbreaks that may be difficult to control. Like other countries in southern Africa, Eswatini depends on South africa for substabtial amounts of its food requirements, including cereals, fruits, vegetables, and meat products. Brands of ready-to-eat cold meat products that were implicated in the listeriosis outbreak in South Africa are also marketed in Eswatini. As a strategy to prevent the spread of the outbreak in Eswatini, the Ministry of Health embarked on a consumer awareness campaign and initiated a recall of affected products. The country had no capacity to test the products to verify presence of Listeria monocytogenes, thereby highlighting the need to strengthen the food control system. This paper reviews the state of the food control system in Eswatini and assesses the readiness of the country to respond to food safety emergencies using the listeriosis outbreak in South Africa as a case study.

Responsiveness to Food Safety Emergencies in Eswatini following the Outbreak of listeriosis in South Africa

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ABSTRACT

The FAO defines food safety as the absence, or safe, acceptable levels, of hazards in food that may harm the health of consumers. Microbiological hazards pose a disproportionate threat to human health in all countries, more so in developing countries due to inadequate resources and fragmented food control systems. The food control system in Eswatini is administered by different departments. The Ministry of Health is responsible for the administration of the Public Health Act, Ministry of Agriculture, the Dairy Act, and the Ministry of Trade, Industry and Commerce is responsible for standards, including food standards. A 2013 Food Safety Bill that aims at coordinating food control activities under a single food control authority has yet to be finalised. The outbreak of listeriosis in South Africa in 2017 revealed the importance of an effective food control system. Under the current food control system, Eswatini runs the risk of food poisoning outbreaks that may be difficult to control. Like other countries in southern Africa, Eswatini depends on South Africa for substantial amounts of its food requirements, including cereals, fruits, vegetables, and meat products. Brands of ready-to-eat cold meat products that were implicated in the listeriosis outbreak in South Africa in 2017 are also marketed in Eswatini. As a strategy to prevent the spread of the outbreak in the country, the Ministry of Health embarked on a consumer awareness campaign and initiated a recall of affected products. The country had no capacity to test the products to verify presence of *Listeria monocytogenes*, thereby highlighting the need to strengthen the food control system in Eswatini. This paper reviews the state of the food control system in Eswatini and assesses the readiness of the country to respond to food safety emergencies, using the listeriosis outbreak in South Africa as a case study.

Keywords: Food safety, listeriosis, Eswatini, Food control, pathogens

Oral Session | Postharvest/Food Technology and Process Engineering

[5-1015-C] Postharvest/Food Technology and Process Engineering (5)

Chair:Akindele Folarin Alonge(University of Uyo, Nigeria)

Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[5-1015-C-01] **THE EFFECT OF DRYING METHODS ON THE QUALITY OF TIGER NUT** *(Cyperus esculentus lativum)*

*Akindele Folarin ALONGE[1], Edikan Ufot GILBERT （1. University of Uyo(Nigeria)）

10:15 AM - 10:30 AM

[5-1015-C-02] **Optimization and Storage Stability Evaluation of Antioxidant Extracts From Batangas Cherry (*Terminalia microcarpa* Decne)**

*Dennis Marvin Opena Santiago[1], Shekayna Eunice Balmes Pacia[1], Jake Lloyd Cabrera Peña[1,2], Claire Solis Zubia[1], Sheba Mae Magbanua Duque[1] （1. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Banos, College, Laguna 4031 Philippines(Philippines), 2. Department of Science and Technology CALABARZON Region, Regional Science and Technology Center Complex, Jamboree Road, Timugan, Los Banos, Laguna 4030 Philippines(Philippines)）

10:30 AM - 10:45 AM

[5-1015-C-03] **Effects of Pre-drying treatment and Drying-air Temperature on Moisture Ratio and Effective Moisture Diffusivity of Tomato (Nigerian Local and Foreign Varieties)**

*Obafemi Ibitayo Obajemihi[1], Joshua Olanrewaju Olaoye[2], Mayowa Saheed Sanusi[1] （1. Food Engineering Department, University of Ilorin(Nigeria), 2. Agricultural and Biosystems Engineering, University of Ilorin(Nigeria)）

10:45 AM - 11:00 AM

[5-1015-C-04] **Extending the Shelf-life of Upland Water Spinach (*Ipomoea aquatica*) Using Trimming, Modified Atmosphere Packaging (MAP) and Low-Temperature Storage**

*Ana Mithuzela Espigol[1], Josephine Agravante[1] （1. Postharvest Horticulture Training and Research Center (PTHRC), College of Agriculture and Food Science (CAFS), University of the Philippines Los Baños (UPLB), Laguna, Philippines(Philippines)）

11:00 AM - 11:15 AM

[5-1015-C-05] **Investigation of Cowpea Variety and Storage Methods on Cowpea Beetle Infestation**

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11:15 AM - 11:30 AM

Oral Session | Postharvest/Food Technology and Process Engineering

[5-1015-C] Postharvest/Food Technology and Process Engineering (5)

Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[5-1015-C-01] THE EFFECT OF DRYING METHODS ON THE QUALITY OF TIGER NUT *(Cyperus esculentus lativum)*

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Keywords: Drying methods, quality, tiger nuts, moisture content, properties

This study aimed at evaluating the effect of different drying methods on the quality of tiger nut (*cyperus esculentum lativum*). Three drying methods: sun drying, oven drying and microwave-oven drying were employed. Analysis of proximate, minerals, anti-nutrient and anti-oxidant composition of fresh (control) and dried tiger nut were carried out using the official method of analysis byt the association of analytical chemist (AOAC, 2010). Fresh tiger nut tubers were divided into four portions. Three of the four portions were dried to constant weight using sun, oven and micro-wave drying methods respectively. The fourth portion of the sample was not dried but serves as the control. Result showed that the proximate composition of fresh and dried tiger nut sample for moisture content ranged from (5-45%), Protein (1.04-3.50%), Ash (0.05-0.51%), fibre (3.69-5.04%). fat (23.48-24.11%). For the dried samples, microwave oven drying had the lowest moisture (5.0%), oven had the highest fibre (3.80%), oven had the highest ash (0.48%), oven had the highest fat (24.11%), and oven had the highest protein (3.50%) contents contents. These values were significantly different from (p<0.05) the control. The minerals composition of the fresh and dried tiger nut ranged from Calcium (1.97mg/g-2.41mg/g), Potassium (2.29mg/g-3,83mg/g), Magnesium (1.03mg/g-5.33mg/g), and Zinc (5.09mg/g-8.11mg/g). Anti-nutrients of dried tiger nut were significantly reduced among other drying methods when compared with the control; anti-nutrient of the fresh and dried tiger nut range from Hydrogen cyanide (HCN) (0.012mg/g-0.401mg/g), Oxalate (0.016mg/g-0.084mg/g). Phytate (0.022mg/g-0.062mg/g), Tannin (Ta) (0.029mg/-0.0364mg/g). Anti-oxidant of the fresh and dried tiger nut ranged from 1, 1-diphenyl-2-picryllydrazyl (DPPH) (0.577%-2.23%), Cupric ion reducing capacity assay (CUPRAC) (0.52%-0.44%), Ferric ion reducing anti-oxidant power assay (FRAP) (0.40%-0.68%). At the end of this study, Oven drying maintained high nutritional content among the drying methods. Microwave oven drying method had the highest retention of its mineral composition when compared with the control. Sun drying had the lowest anti-nutrient among the drying methods. Microwave oven drying was effective in its anti-oxidant activity with reference to the control.

The Effect of Drying Methods on the Quality of Tiger nut
(Cyperus esculentus lativum)

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ABSTRACT

This project aimed at evaluating the effect of different drying methods on the quality of tiger nut (*cyperus esculentum lativum*). Three drying methods: sun drying, oven drying and microwave-oven drying were employed. Analysis of proximate, minerals, anti-nutrient and anti-oxidant composition of fresh (control) and dried tiger nut were carried out using the official method of analysis by the association of analytical chemist (AOAC, 2010). Fresh tiger nut tubers were divided into four portions. Three of the four portions were dried to constant weights using sun, oven and micro-wave drying methods respectively. The fourth portion of the sample was not dried but serves as the control. result showed that the proximate composition of fresh and dried tiger nut sample for Moisture content ranged from (5-45%), Protein (1.04-3.50%), Ash (0.05-0.51%), fibre (3.69-5.04%). fat (23.48-24.11%). for the dried samples, microwave oven drying had the lowest moisture (5.0%), oven had the highest fibre (3.80%), oven had the highest ash (0.48%), oven had the highest fat (24.11%), and oven had the highest protein (3.50%) contents. These values were significantly different from ($p<0.05$) the control. The minerals composition of the fresh and dried tiger nut ranged from calcium (1.97mg/g-2.41mg/g), Potassium (2.29mg/g-3,83mg/g), Magnesium (1.03mg/g-5.33mg/g), and Zinc (5.09mg/g-8.11mg/g). Anti-nutrients of dried tiger nut were significantly reduced among other drying methods when compared with the control; anti-nutrient of the fresh and dried tiger nut range from hydrogen cyanide (HCN) (0.012mg/g-0.401mg/g), oxalate (0.016mg/g-0.084mg/g). Phytate (0.022mg/g-0.062mg/g), Tannin (Ta) (0.029mg/-0.0364mg/g). anti-oxidant of the fresh and dried tiger nut ranged from 1, 1-diphenyl-2-picryllydrazyl (DPPH) (0.577%-2.23%), Cupric ion reducing capacity assay (CUPRAC) (0.52%-0.44%), Ferric ion reducing anti-oxidant power assay (FRAP) (0.40%-0.68%). At the end of this study, oven drying maintained high nutritional content among the drying methods. Microwave oven drying method had the highest retention of its mineral composition when compared with the control. Sun drying had the lowest anti-nutrient among the drying methods. Microwave oven drying was effective in its anti-oxidant activity with reference to the control.

Keywords: Drying, Tiger nuts, Sun drying, Oven drying, Microwave drying, Drying rates, Drying methods

1. INTRODUCTION

Tiger nut "*Cyperus esculentus lativum*" is an underutilized tuber of family Cyperaceae, it produces rhizomes from the base of the tuber that is spherical (Devries and Feuke, 1999). It is a tuber that grows freely and is consumed widely in Nigeria and other parts of West Africa.
Tiger nuts exist in varieties (black, brown and yellow which are cultivated. Among these, the yellow variety is preferred over others because of its inherent properties such as large size, attractive color and fleshier nature. It yield more milk upon extraction, contains lower fat and higher protein (Okafor and Okolo, 2003). Tiger nut tubers appear long or round in shape with a dimension of 8mm to 16mm, the smaller size however, are not used for human consumption. Recently, there is awareness for increased consumption of tiger nut (Belewu and Abodunrin, 2006; Belewu, 2007). When hydrated, it is slightly harder (nut texture), but with a rather more intense and concentrated taste. The cultivation time is April to November.
Tiger nut, a tuber with sweet and nutty taste can be consumed raw, roasted, dried or as tiger nut milk or oil (Rita,2009) .It can be stored and rehydrated by soaking without losing the crop texture

which ensures acceptable sensory quality (Tucson, 2003). Drying of agricultural products helps to reduce the moisture content to a level that halts or control microbial growth and to reduce deteriorative chemical reaction in order to extend the shelf life of food (Mujumdar and Law, 2010).

In most agricultural based economies like Nigeria, large quantities of food products are dried to improve shelf life, reduce packaging costs, lower weights, enhance appearance, retain original flavor and most importantly maintain nutritional quality (Baysal *et al.*, 2003; Demir *et al.*, 2007; Simal *et al.*, 2000; Ertekin and Yaldiz, 2004).

Sun, oven and microwave oven drying are common drying methods for agricultural crops. These drying methods have been reported to affect the nutrient composition of food in various ways. It can either increase the concentration of some nutrients by making them more available or decrease the concentration of some nutrients (Hassan *et al.*, 2007; Morris *et al.*, 2004; Ladan *et al.*, 1997). Therefore, this project seeks to investigate the effect of different drying methods like sun drying, oven drying, and microwave oven drying on the quality of tiger nuts.

2. MATERIALS AND METHODS

SAMPLES COLLECTION AND PREPARATIONS

Fresh Tiger nuts were purchased from Itam Main Market, Uyo. Akwa Ibom State, Nigeria. The tubers were thoroughly screened to remove the bad ones and stones. They were washed, air dried and divided into four portions. Three of the four portions were dried to constant weight using sun, oven and micro-wave drying methods respectively. The fourth portion was not dried but was used as fresh sample which served as the control

Control: Hundred grammes of fresh tiger nut sample were kept as the control to be compared with the dried tiger nut samples.
Sun Drying: Hundred grammes of the samples were kept in the sun between 10:30 am to 3:30 pm daily and were dried to constant weight (22.690 g) for 60 hours.
Oven Drying: Hundred grammes of the samples was also placed in an electric oven and dried to constant weight (27.328 g) at 65°C for 20 hours.
Microwave Oven Drying: Hundred grammes of the samples were dried using a microwave oven to constant weight (28.120 g) for at 50°C for 15 minutes.

2.1 Materials and Equipment

For a successful execution of this research work, Sulphuric acid (H_2SO_4), copper sulphate ($CUSO_4$), sodium sulphate (Na_2SO_4), boric acid (H_3BO_3), hexane (C_6H_{14}), sodium hydroxide (NaOH) would be used in carrying out the proximate analysis of the samples.
Equipment to be used include: Kjeldahl (soxhlet) apparatus, water bath, electric oven (model PVHB-90G2HA), fume cupboard, desiccators, crucibles, Buckner funnel, measuring scale, muffle furnace (by Uhlg, Kern, U.S.A), sifter, JENWAY 6100 Spectrophotometer, Pearson Gallenkamp Flame analyzer, Buch Model 205 Atomic Absorption Spectrophotometer, electric oven, micro wave oven (Westpoint Microwave oven dryer), Digital thermometer, Weighing Balance, conical flask (250ml), volumetric flask, reflux device, acid burette, filtration device etc were used in this study.

2.2 Proximate Analysis of Tiger Nuts

The proximate components of the fresh, dried tubers of tiger nuts were using the standard methods of Analysis of Association of Official Chemists (AOAC), 2010. Crude protein, crude lipid, carbohydrate, Moisture, and Ash contents in the samples was analyzed. The methods are described below. The same procedures were carried out on all samples.

2.3 Mineral Content Analyses of Tiger Nuts

The minerals to be analysed would be Potassium, Calcium, Magnesium and Zinc. Potassium would be determined using Gallenkamp Flame analyzer, while calcium, magnesium and zinc would be determined using the atomic absorption spectrophotometer (model Unicam 900, Buck Scientific).
The digest solutions of the samples were prepared by weighing 1 grams of each of the powdered plant samples, these were digested with aqua regia at 130ºC using electric hotplate for 30 minute .the

filtered was made up to 100ml after filtration using 100ml volumetric flask. Standard solutions of the metal to be analyzed were prepared. The atomic absorption spectrophotometer (model Unicam 900, Buck Scientific) was set with power on for ten minutes. The standard minerals solutions were injected to calibrate the AAS using acetylene gas. An aliquot of ash solutions were injected and the concentrations obtained from the AAS.

Two grammes (2 g) of each tiger nut sample would be heated gently over a Bunsen burner flame until most of the organic matter was destroyed. This will be further heated strongly in a muffle furnace for several hours until white- grey ash was obtained. The ash material was cooled. About 20 ml of distilled water and 10 ml of the dilute hydrochloric acid was added to the ashen material. This mixture would be boiled, filtered into a 250 ml volumetric flask, washed thoroughly with hot water, cooled and made up to volume.

2.4 Anti-nutrients Analysis

2.4.1 Hydrogen Cyanide

Extraction of hydrogen cyanide was done using Wang and filled method. The sample (2g) was ground into paste and dissolved in distilled water (50ml) using a conical corked flask. The extract was allowed to stay overnight and the filtered solution was used for the cyanide determination. Alkaline picrate 4ml was added to 1m of the filtrate in a corked test tube and incubated in water bath for 15minutes. Reddish colour developed and the absorbance was taken using a spectrometer at 490nm (AOAC, 1984). Also, the absorbance of the blank containing only 1ml distilled water and 4ml alkaline picrate solution was taken and the extrapolation of the cyanide content from the cyanide standard curve.

Concentration of hydrogen cyanide is thus as follows;

$$\frac{absorbance\ test\ x\ conc.standard}{absorbance\ of\ standard\ x\ weight\ of\ sample} x \frac{100}{1} \qquad (1)$$

2.4.2 Determination of Oxalate by Titration Method

The oxalate content of the sample was determined using titration method. It involves three general steps which include digestion, precipitation and $KMnO_4$ titration.

Digestion: 5g of the sample was introduced into a 250ml beaker suspended in 95ml of distilled water and 5ml 6N HCl was added to the beaker. The mixture was heated on a water bath at 50^0C for 2 hours. The digestion was filtered and diluted with distilled water to 126ml.

Precipitation: 50ml of the filtrate was placed in a 100ml beaker and drops of methyl red indicator was added which evaporated on eating to 250ml in volume. The sample was filtered to remove the precipitate containing ferrous irons. The filtrates were again treated with 5ml NH_4OH and heated to 90^0C and 10mn of 5% CaCl solution was added and stirred constantly as heat was applied and allowed to cool overnight at 5^0C. The solution was then centrifuged (filtered) at 2500rpm for 5 minutes. The supernatant was decanted and he precipitate were obtained which was washed into a beaker with H_2SO_4 (10ml of 20% v/v) and diluted with 125ml of distilled water.

Titration: the 125ml aliquot solution was heated near boiling point (90^0C) and was titrated against 0.05N standardized $KMnO_4$ solution to a faint pink color which persists for 10seconds.
The calcium oxalate content is calculated using the formular 0.05N $KMnO_4$ = 2.2g Oxalate.

2.4.3 Determination of Phytate

The phytate content of the tiger nut was determined by Maga method. Two (2g) grammes each finely ground flour sample was soak in 20ml of 0.2N HCl and filtered. After filtration, 0.5ml of the filtrate was mixed with 1ml ferric ammonium sulphate solution in a test tube, boiled for 30min in a water bath, cooled in ice for 15minute and centrifuged at 3000× g for 15 minutes. One millitre of the supernatant was mixed with 1.5ml of 2,2-pyridine solution and the absorbance measured in a spectrophotometer at 519nm. The concentration of phytic acid was obtained by extrapolation from a standard curve using standard phytic acid solution.

2.4.4 Determination of Tannin Content
For Tannin determination, 10ml 70% aqueous acetone was added to 2g of finely ground sample in a bottle and properly covered. The bottle was put in an ice bath shaker for 2h at 30oC. The solution was then centrifuged and the supernatant stored in ice. From the supernatant, 0.2ml was pipette into 0.8ml of distilled water. Standard tannic acid solution was prepared. Folin reagent (0.5ml) was added to both sample and standard followed by 2.5ml 20% Na_2CO_3. The solution was vortexed and allowed to incubate for 40minute at room temperature after which the absorbance was read at 725nm. The concentration of tannin in the sample was estimated from the standard tannic acid curve.

2.5 Antioxidants Analysis

2.5.1 DPPH Radical Scavenging Assay: The free radical scavenging capacity of the extracts from different plant samples were estimated according to Baraca, 2003 with slight modification using the stable 1,1-diphenyl-2-piccryllydrazyl (DPPH) radical which has an absorption maximum at 515nm. A solution of the radical is prepared by dissolving 2.4mg DPPH in 100ml methanol. A test solution100-500Nl was added to 3.95ml (4ml) of methanolic DPPH. The mixture was shaken vigorously and kept at room temperature for 30min in the dark. Absorbance of the reaction mixture was measured at 515nm spectrophotometric absorbance of the DPPH radical without antioxidant i.e. blank was also measured. All the determinations were performed in duplicates. The capacity to scavenge the DPPH radical was calculated using the following equation:

DPPH Scavenged (%) $= \dfrac{(AB - AA)}{AB * 100}$ $\hspace{3cm}$ (2)

Where AB = Absorbance of Blank
AA is absorbance of the antioxidant at t = 30minutes

2.5.2 Ferric Ion Reducing Antioxidant Power Assay (Frap)
Ferric ion reducing power was measured according to the method of Oyaizu with a slightest modification.
Procedure: Hydroalcoholic extract of the sample in different concentration ranging from 100nl to 500nl were mixed with a 2.5mM phosphate buffer and 2.5ml, 1%, w/v potassium ferric cyanide, and then the mixture was I incubated at 50*c for 30minutes. Afterward, 2.5ml of 10%, w/v trichloroacetic acid and 0.5ml 0.1%, w/v ferric chloride were added to the mixture, which was kept aside for 10min. finally, the absorbance was measured at 700nm. Ascorbic acid was used as positive reference standard. All assays were run in duplicates and averaged.

2.5.3 Cupric Ion Reducing Capacity Assay (Cuprac)
Cupric ion reducing capacity was measured in accordance to the method of Apal.
Procedure
1ml, 10mM cupric chloride, 1ml 7.5mM neocuproine and 1ml, 1M ammonium acetatate buffer of PH 7 solutions were to test tubes containing 2ml of distilled water. Hydroalcoholic extract of the sample in different concentration ranging from 100nl to 500nl were added to each test tube separately. These mixtures were incubated for half an hour at room temperature and measured against blank at 450nm. Ascorbic acid was used as positive reference standard. All methods were repeated in duplicates in order to get a mean value.

2.6 Statistical Analysis
The experiments were conducted in duplicates. The mean and standard deviation of the result data from the experiment will be calculated and analyze using single factor ANOVA in the Statistical Package for Social Science (SPSS, 2017) Software (SPSS version 20 for windows). The Duncan's New Multiple Range Test (DNMRT) and Ordinary Least Significant Difference (LSD) were also used to determine the significant difference between mean values (Spiegiel *et al*., 2008).

3. RESULTS AND DISCUSSIONS

3.1 Drying Rate Curve

Below are the drying rate curves showing different drying methods with different drying rates. Figure 4.1 shows the drying rate curve for sun drying. Here, there was a rapid increase in drying rate from 0 - 14.44 gH_2O/hr between 0-5 hours. A rapid decrease in drying rate from 14.44 - 0.177gH_2O/hr between 5 - 45 hours of drying time, and a minimum of 0.114 gH_2O/hr constant drying rate was found between 45-60 hours of drying time. Figure 4.2 shows the drying rate curve for oven drying. Here, there was a rapid increase in drying rate from 0-1 hour with a maximum corresponding drying rate of 66.311gH_2O/hr. Between 1-12 hours of drying time, there was a rapid decrease in drying rate from 66.311 - 0.851gH_2O/hr and between 12-20 hours of drying, the drying rate decreased from 0.851 gH_2O/hr to a constant value of 0.237 gH_2O/hr. Figure 4.3 shows the drying rate curve for microwave oven drying. Here, there was a rapid increase in drying rate from 0 - 11.058 gH_2O/min between 0-5 minutes and decreased from 11.058 - 0.662gH_2O/min between 5-15 minutes of drying time. Different drying methods had varying energy output and usage and these had different impact on the samples; and also affect the quality of product differently. Generally, the drying rate decreased as the drying time increased. For sun drying, the drying rate was low and it took about 60 hours to dry to a bone dry weight of 22.690 grams. For oven drying, the drying rate was faster compared with the sun drying, and it took about 20 hours to dry to a bone dry weight of 27.328 grams. Microwave oven had the highest drying rate, which took about 15 minutes to dry to a bone dry weight of 28.120 grams. Microwave oven had the highest drying rate among other drying methods.
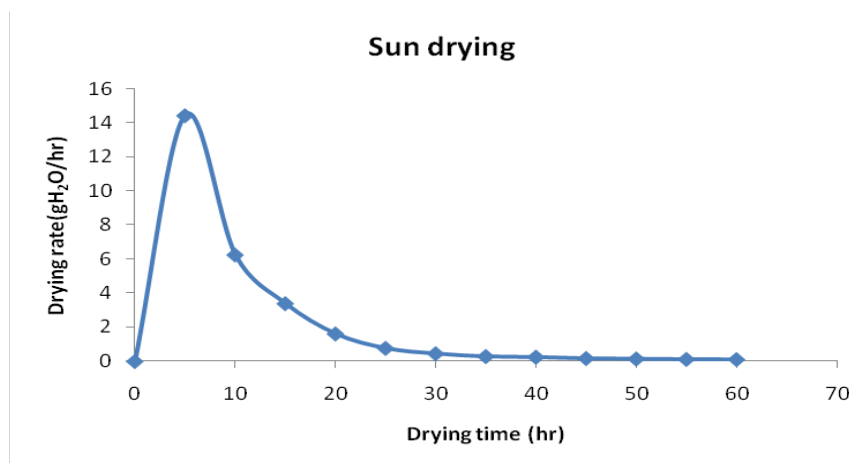
Figure 3.1: Drying rate curve for sun drying.
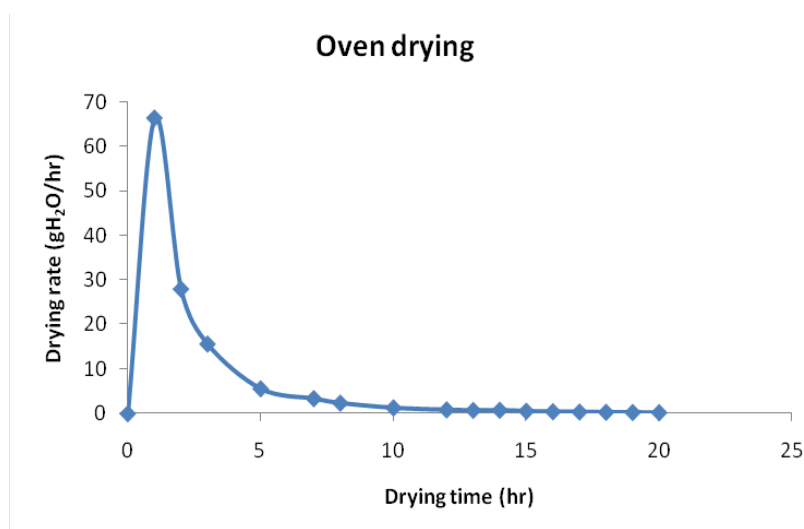
Figure 3.2: Drying rate curve for Oven drying.
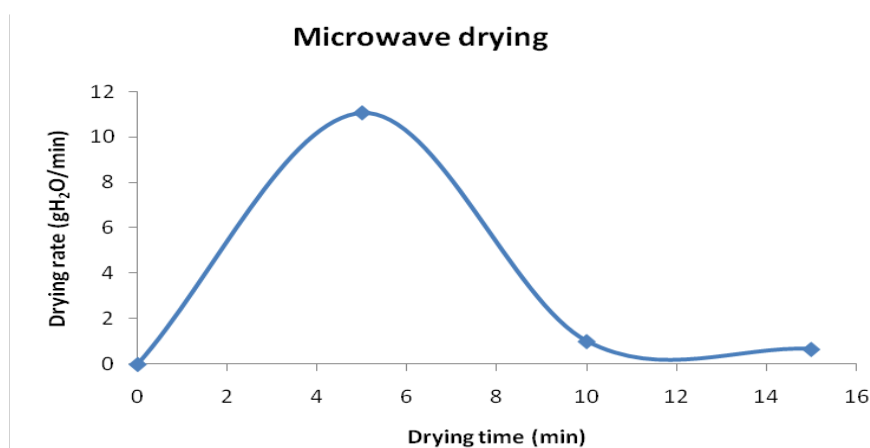
Figure 3.3: Drying rate curve for Microwave oven drying.

3.2   Effect of drying methods on the proximate compositions of tiger nut

Table 3.2A: Effect of different drying methods on proximate composition of tiger nut

Drying Method	Moisture Content (%)	Crude Fibre (%)	Ash Content	Crude Lipid
Fresh	$45.0050 \pm 0.00707^{*}$	$5.0495 \pm 0.07000^{*}$	$0.5105 \pm 0.00071*$	$23.4830 \pm 0.00141^{*}$
Microwave	$5.0005 \pm 0.0071^{*}$	3.6985 ± 0.00071^{b}	0.0495 ± 0.00071^{P}	$24.1075 \pm 0.00071^{*}$
Oven	10.0050 ± 0.00707^{a}	$3.8050 \pm 0.00707^{*}$	$0.4805 \pm 0.00071^{*}$	$24.1150 \pm 0.00424^{*}$
Sun	10.0010 ± 0.00141^{a}	3.6910 ± 0.00141^{b}	$.0505 \pm 0.00071^{P}$	$24.0340 \pm 0.00141^{*}$

Values are means $\pm$ standard deviation from duplicate analyses
Values with asterisk (*) showed significance difference in their mean at 5 % level. Values with same alphabet in the same column did not differ in their mean at 5% level of significance.

Table 3.2B: Effect of different drying methods on proximate composition of tiger nut

Drying Method	Crude protein	Total Carbohydrate (%)	Caloric value (Kcal)
Fresh	1.7600± 0.01414*	69.1965 ± 0.08697*	495.1750± 0.27577*
Microwave	1.7005±1.7005*	70.4435 ± 0.00212*	505.5455±0.00354[h]
Oven	3.5035 ± 0.00495*	70.0955 ± 0.00778*	503.4310 ± 0.04950*
Sun	1.0400 ± 0.01414*	71.1835 ± 0.01020*	505.2000 ± 0.00424[h]

Values are means ± standard deviation from duplicate analyses
Values with asterisk (*) showed significance difference in their mean at 5 % level of   significance. Values with same alphabet in the same column did not differ in their mean

Tables 3.2A and B present the effect of different drying methods on the proximate composition of fresh and dried tiger nuts per 100 grams. The moisture content of tiger nut ranged from 45% in the fresh sample (control) to 5.0% in the microwave oven; only oven and sun drying methods did not produce significant difference in their mean values, since p [0.474] > 0.05. Reduction in the moisture content as observed in this study decreases the perishability of tiger nut, adds value and also extends the shelf life, thereby making it available throughout the year, similar to the report of Demirel and Turhan (2003) and Emperatriz *et al.* (2008) .The tiger nut samples were samples were significantly different (p<0.05) in fibre content except for microwave and sun drying methods which did not produce significant difference in their mean values of fibre content since p [0.842] > 0.05. The fresh tiger nut sample had higher (5.04%) fibre content than the dried samples as compared with (Okorie and Nwanekezi, 2014).The reduction observed in the dried sample might be due to the fact that drying softens cellulose, and encourage loss of indigestible plant components, causing the cells to separate easily and making the nut easier to digest (Cameron, 1983). Loss of soluble fibre by hydrolysis, enzymatic degradation and decomposition caused fibre to reduce (Morris *et al.,* 2004). Ash content was highest in the fresh sample (0.51%) and it was lowest in the microwave dried tiger nut (0.04%) but only microwave and sun drying methods did not produce significant difference in their mean values, since p [0.230] > 0.05, this is as a result of their moisture content and leaching of it minerals during drying as reported by Ogunlade *et al.* (2015). There was a significant increase in lipid content as fresh sample had the lowest lipid content (23.4%) but highest in the oven dried sample (24.1%). Increased in the lipid content of dried tiger nuts is attributed to concentration of fat due to moisture loss (Ndubuisi, 2009). Protein content decreases significantly when compared with the fresh sample, this is in line with the report by Miroslawaet *et al.*(1997) that heat application caused the unzipping of the hydrophobic force leading to partial or complete disruption of the primary, secondary tertiary or quaternary structure of protein molecules thereby leading to the protein content of the dried sample. Crude protein was lowest in the sun dried sample (1.04%) when compared with that of the fresh sample (1.76%). There was an increased in the carbohydrate contents when compared with the fresh sample (69.19%), this may be attributed to moisture loss which leads to concentration of nutrient (Ndubuisi, 2009); Total carbohydrates were highest in the sun dried tiger nuts (71.1%) but lowest in the oven dried samples (70.09%). Caloric value increased significantly among the drying methods with reference to the control. Microwave dried tiger nut had the highest caloric value (505.5 KJ) and lowest in the fresh sample (495.17%) ; Only microwave and sun drying methods did not produce significant difference their mean values since their p [0.069] > 0.05.

3.3 Effect of Drying Methods on the Minerals Composition of Tiger Nut

Table 3.3: Mineral composition of fresh and dried tiger nuts (mg/100g)

Treatment		Calcium	Potassium	Magnesium	Zinc
	Fresh	2.4150±0.00141*	3.8305±0.0071*	5.3305±0.7566*	8.1165±0.0071*
	Microwave	2.0845±0.00071[a]	2.3050±0.00141*	2.0320±0.141*	6.0080±0.00141*
	Oven	2.0845±0.00071[a]	2.2960±0.00141[b]	1.2090±0.000*	5.1210±0.00141*
	Sun	1.9740±0.00141*	2.2960±0.00141[b]	1.0375±0.00071*	5.0920±0.00141*

Values are means ± standard deviation from duplicate analyses
Values with asterisk (*) showed significance difference in their mean at 5 % level of   significance. Values with same alphabet in the same column did not differ in their mean

Table 3.3 present the effect of different drying methods on the mineral composition of tiger nuts. The minerals composition of the dried tiger nuts was reduced when compared with the fresh sample. The fresh sample indicates high calcium content (2.41mg) but lowest in sun dried sample (1.97mg) when compared with the other drying methods. Calcium content of the micro wave oven and the oven dried tiger nut were not significantly different with their means at 5% level of significance. Potassium content was highest in the fresh sample (3.83mg) but lowest in oven and sun dried tiger nuts (2.29mg) which were not significantly different with their mean. There was a significant difference among Magnesium content of tiger nuts with its content highest in the fresh sample (5.33mg) but lowest in the sun dried samples (1.03mg). Fresh tiger nut had the highest Zinc composition (8.11mg) but lowest in the sun dried tiger nuts (5.09mg).The decrease in the mineral content of tiger nuts after drying, suggest that the presence of anti-nutritional factors such as oxalate and phytate in this tuber made these minerals unavailable by reacting with them, this is similar to the report of Akpan and Umoh (2004). Microwave oven drying had the highest mineral retention when compared with other drying methods with reference to the control sample.

3.4 Effects of drying methods on the anti nutrient composition of tiger nuts

Table 3.4 Anti nutrient composition of fresh and dried tiger nuts (mg/100g)

Treatment	HCN	Oxalate	Phytate	Tannin
Fresh	0.0122±0.00007*	0.084±0.00141*	0.0622±0.0014*	0.0292±0.0021*
Microwav	0.0226±0.00078*	0.1410±0.00141*	0.0482±0.00141*	0.314±0.00014*
Oven	0.0279±0.00007*	0.0435±0.00212*	0.0227±0.0028[a]	0.0358±0.000*
Sun	0.4011±0.0007*	0.0160±0.00141*	0.0227±0.00028[a]	0.0364±0.00035*

Values with asterisk (*) show significance difference in their means at 5% level of significance.
Values with same alphabet in the same column did not differ in their mean.

Table 3.4 present the effects of drying methods on the anti nutrient composition of fresh and tiger nut. It shows that drying methods had a significant reducing effect on anti-nutrients compositions except for Hydrogen cyanide and Tannin. Reduction in anti-nutrient of the sample was observed mostly in sun dried sample; this is as a result of the evaporation of toxic chemicals from tiger nut samples during sun drying into the atmosphere. Oxalate content was reduce significantly when compared with the fresh sample, it was highest in the microwave sample (0.141mg) and lowest in the sun sample (0.016mg). Phytate was highest in the fresh sample (0.0622mg) and lowest in the oven and sun dried samples (0.0227mg); Hydrogen Cyanide content of the tiger nut were increase and significantly different with their means at 5% level of significance. It was lowest in the fresh sample (0.0122mg) and highest in the sun dried samples (0.4011mg). Tannin indicates a high content in micro wave oven sample (0.314mg) but low in the fresh sample (0.029mg).

3.5 EFFECT OF DRYING METHODS ON ANTI OXIDANTS OF FRESH AND DRIED TIGER NUTS

Table 3.5 Effect of drying methods on anti oxidants of fresh and dried tiger nuts (mg/100g)

Treatment	DPPH	Cuprac	FRAP
Fresh	2.2315±0.00212*	0.0520±0.0000a	0.4040±0.00283*
Microwave	1.0400±0.00141*	0.4450±0.0000*	0.6820±0.00141*
Oven	0.9710±0.00283*	0.2890±0.0000*	0.4065±0.00212*
Sun	0.5775±0.0.00212*	0.0530±0.00141[a]	0.5880±0.00141*

Values with asterisk (*) show significance difference in their means
Values with same alphabet in the same column did not differ in their mean.

Table 3.5 highlights the effect of drying methods on the antioxidant activity of fresh and dried tiger nuts. Their antioxidant activity was significantly different at 0.05 significant level among their mean. DPPH was found to be highest in the fresh sample (2.23mg) but lowest in the sun dried tiger nuts (0.577mg). Cupric ion reducing capacity assay CUPRAC was highest in the microwave sample (0.445mg) but lowest in the fresh sample (0.052mg); only sun drying method and fresh did not produce significant difference their mean values since p [0.184] > 0.05.Ferric ion reducing antioxidant power assay (FRAP) was highest in the microwave sample (0.682mg) but lowest in the fresh sample (0.404mg). Microwave dried sample was highest in its anti-oxidants activity to neutralize the toxicity of anti-nutrients in tiger nuts.

4. CONCLUSION

Preservation of food by drying is a common practice in different parts of the world and it is used to extend the shelf life of food. Drying allows food to be preserved by removing the moisture in the food, in order to prevent the growth of microorganisms that cause deterioration (Mukhtar, 2009). It ensures their availability all year round, reduce post harvest losses and achieve food security. In this study, drying methods used includes: sun drying, oven drying and microwave oven used were capable of preserving the nutrients in the food crops without total loss of any nutrient. The following conclusions were deduced:

a. Oven and micro wave drying were observed to be more hygienic and faster than the sun drying. However, Micro wave drying had the highest drying rate than oven drying and it also gave the lowest **moisture content** in this study, suggesting a higher capacity to prevent microbial growth and decay in the dried samples, thus, confers a greater increase in shelf life on the dried samples.

b. There were decrease in fiber, ash and protein contents of dried samples, using all the drying methods whiles fat, carbohydrate and energy value were increased.

c. The drying methods had a reducing effect on the minerals composition of the dried tiger nuts when compared with the fresh sample, though microwave samples had the highest retentions among the other drying methods.

d. This study showed that drying method reduced the anti-nutrients in tiger nut (*Cyperus esculentus lativum) w*hen compared with the fresh sample.

e. There was a significant difference in the anti-oxidant activity of the tiger nut tubers. Cupric ion reducing capacity (CUPRAC) and ferric ion reducing antioxidant power (FRAP) increased with all the drying methods.

f. The drying time affected the anti-oxidant activity of the product

g. At the end of this study, it was observed that oven drying had the best nutritional composition, micro wave dried tiger nuts had the highest anti-oxidant activity and minerals composition. Sun drying had the lowest anti nutrient composition on tiger nutsas a result of decomposition of these anti nutrients into the soil and escape into the atmosphere during sun drying process.

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10:30 AM - 10:45 AM (Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room C)

[5-1015-C-02] Optimization and Storage Stability Evaluation of Antioxidant Extracts From Batangas Cherry (*Terminalia microcarpa* Decne)

*Dennis Marvin Opena Santiago[1], Shekayna Eunice Balmes Pacia[1], Jake Lloyd Cabrera Peña[1,2], Claire Solis Zubia[1], Sheba Mae Magbanua Duque[1]   (1. Institute of Food Science and Technology, College of Agriculture and Food Science, University of the Philippines Los Banos, College, Laguna 4031 Philippines(Philippines), 2. Department of Science and Technology CALABARZON Region, Regional Science and Technology Center Complex, Jamboree Road, Timugan, Los Banos, Laguna 4030 Philippines(Philippines))
Keywords: Solvent extraction, Box-Behnken design, Batangas Cherry, DPPH scavenging activity, Anti-microbial activity

Effects of extraction parameters, including temperature, solvent to sample (S/S) ratio and ethanol concentration on % 2,2-Diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity were optimized using Box-Behnken design (BBD) of experiment. Moreover, effects of pH (3.5 to 9), and storage condition (5 and 25 ^{o}C) on the stability and antimicrobial activity of antioxidant extracts from Batangas Cherry were determined. Result showed that the optimum condition for extraction antioxidants from Batangas Cherry was at 80°C, 10mL g^{-1} S/S and 51.66% ethanol. Batangas cherry extracts exposed at pH8 and 9 showed significant decrease in antioxidant and antimicrobial activities. On the other hand, storage at 5°C better retained the antioxidant and antimicrobial activities of Batangas Cherry extracts. The baseline data in this research is important on maximizing the potential of Batangas as source of functional ingredient for food processing.

Oral Session | Postharvest/Food Technology and Process Engineering
[5-1015-C] Postharvest/Food Technology and Process Engineering (5)
Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[5-1015-C-03] Effects of Pre-drying treatment and Drying-air Temperature on Moisture Ratio and Effective Moisture Diffusivity of Tomato (Nigerian Local and Foreign Varieties)

*Obafemi Ibitayo Obajemihi[1], Joshua Olanrewaju Olaoye[2], Mayowa Saheed Sanusi[1] （1. Food Engineering Department, University of Ilorin(Nigeria), 2. Agricultural and Biosystems Engineering, University of Ilorin(Nigeria)）
Keywords: Hausa, Tiwantiwa, Honey and Sugar, Tomato

Tomato is a crop that is highly perishable and there are huge postharvest losses incurred annually in Nigeria. Drying of the fruit is important and suitable for developing economies. However, the heat employed during drying of tomato can influence its quality adversely, as a result it is important to use pre-drying treatments prior to drying operation. Therefore, the aim of this research was focussed on studying the effects of pre-drying treatment and drying-air temperature on moisture ratio (MR) and effective moisture diffusivity of tomato. Three varieties (Hausa, Tiwantiwa and Roma VFN) of fresh tomato were obtained from local farmers in Ilorin province, Kwara state of Nigeria. The samples were sorted and cleaned under running water, and were sliced at different thicknesses (5 mm, 7.5 mm and 10 mm), deseeded and blanched in hot distilled water at 90oC. The samples were further pre-treated using different chemical and osmotic solutions (2% ethyl acetate, 1% MgCl2 .6H_2O and 0.5% $Na_2S_2O_5$, 0.5% NaCl and 40oBx of honey and sugar) and Control (Non-pretreated). Samples were drained for 10 minutes after pre-drying treatment and were dried at different temperatures (45, 55 and 65oC) in an automated forced convection cabinet dryer (FCCD) instrumented for the purpose of this experiment. Weight loss of the samples were recorded at different intervals (15 - 60 min) on the trays per stage with the aid of a weight reduction sensing mechanism attached through the rear of the dryer. The drying process was stopped (through a computer system connected to the dryer) when the samples had reached their final moisture content <5% (db). The data obtained from the drying process were used to compute the samples moisture ratio and effective moisture diffusivity and were analyzed using regression and analysis of variance (ANOVA) with Design expert v. 6.0.6 statistical tool at p <0.05. The results obtained show that samples lowest MR were obtained under these conditions; processed Hausa variety, 10 mm thickness, ethyl acetate pre-drying treatment and dried at 55oC while highest effective moisture diffusivity were obtained under these conditions processed Hausa variety, 5 mm thickness, honey and sugar pre-drying treatment and dried at 65oC in a FCCD. It was therefore concluded that processed Hausa variety was more preferable to other varieties used as it promotes low MR and high effective moisture diffusivity during the drying process. This will help reduce energy consumption associated with drying process.

Effects of Pre-drying Treatment and Drying-air Temperature on Moisture Ratio and Effective Moisture Diffusivity of Tomato (Nigerian Local and Foreign Varieties)

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ABSTRACT

Tomato is a crop that is highly perishable and there are huge postharvest losses incurred annually in Nigeria. Drying of the fruit is important and suitable for developing economies. However, the heat employed during drying of tomato can influence its quality adversely, as a result it is important to use pretreatments prior to drying operation. Therefore, the aim of this research was focused on studying the effects of pretreatment and drying-air temperature on moisture ratio (MR) and effective moisture diffusivity of tomato. Three varieties (*Hausa, Tiwantiwa* and Roma *VFN*) of fresh tomato were obtained from local farmers in Ilorin province, Kwara state of Nigeria, the samples were sorted and cleaned under running water, and were sliced at different thicknesses (5 mm, 7.5 mm and 10 mm), deseeded and blanched in hot distilled water at $90^{\circ}C$. The samples were further pretreated using different chemical and osmotic solutions (2% Ethyl acetate, 1% $MgCl_2 \cdot 6H_2O$ and 0.5% $Na_2S_2O_5$, 0.5% NaCl and $40^{\circ}Bx$ of Honey and Sugar) and Control (Non-pretreated). Samples were drained for 10 minutes after pre-drying treatment and were dried at different temperatures (45, 55 and $65^{\circ}C$) in an automated forced convection cabinet dryer (FCCD) designed for the purpose of this experiment. Moisture loss of the samples were recorded at different intervals (15 - 60 min) on the trays per stage with the aid of a weight loss sensing mechanism attached through the rear of the dryer. The drying process was stopped (through a computer system connected to the dryer) when the samples had reached their final moisture content < 5% (db). The data obtained from the drying process were used to compute the samples moisture ratio and effective moisture diffusivity and were analyzed using regression and analysis of variance (ANOVA) with Design Expert V 6.0.6 statistical tool at $p < 0.05$. The results obtained show that samples lowest MR were obtained under these conditions; processed *Hausa* variety, 10 mm thickness, Ethyl acetate pre-drying treatment and dried at $55^{\circ}C$ while highest effective moisture diffusivity were obtained under these conditions processed *Hausa* variety, 5 mm thickness, Honey and Sugar pre-drying treatment and dried at $65^{\circ}C$ in a FCCD. It was therefore concluded that processed *Hausa* variety is more preferable to other varieties used as it promotes low MR and high effective moisture diffusivity during the drying process. This will help reduce energy consumption associated with drying process.

Keywords: Honey and Sugar Hausa Tiwantiwa Slice thickness Tomato

1. INTRODUCTION

Tomato is one of the major vegetable crops cultivated in Nigeria and has been known to be highly perishable (Onifade et al., 2013; Idah and Obajemihi, 2014). Drying of fruits and vegetables such as tomato is gaining popularity in Nigeria, where post-harvest loss of farm produce is on increase on yearly basis, due to poor post-harvest handling techniques; as a result, Nigerians have spent a whooping sum of $1bn annually on imported tomato products (UNEP, 2016). Drying is a heat and mass transfer phenomenon and has saved more than 20% of crops that are perishable in the world; by extending their shelf lives and ensuring food security (Sohail et al., 2011). Drying is important and most times indispensable in the formulation of functional food products (Trivedi et al., 2011). Drying

of tomato products usually occur in the falling rate period as the moisture content tends to decrease with time.

It is important to study and understand mass transport mechanisms such as moisture ratio (MR) and effective moisture diffusivity (D_{eff}) responsible for drying of tomato and what to be done during pre-drying and drying processes to favour them. Moisture ratio is the ratio of the instantaneous moisture content to that of the fruit's initial moisture content. Effective moisture diffusivity is an internal transport phenomena and it is the rate at which moisture is moved from the center of the fruit to its surface where it will be evaporated (Onwude et al., 2016). D_{eff} is a function of drying-air temperature and samples' MR and was seen as an important mass transport mechanism when it comes to studying drying processes involving fruit and vegetable (Onwude et al., 2016). Zogzas et al. (1996), stated that increase in temperature brings about increase in effective diffusivity but changes with respect to moisture content. When the temperature of food is high, the water molecules in it are bounded loosely to food matrix than at low temperature, therefore more energy is required to remove moisture at lower temperatures compared with high temperatures. Also food structure and void fraction present can significantly affect moisture diffusivity and hence reported that at low porosity, value of effective diffusivity of moisture is majorly by liquid diffusion which is different from that obtainable for granular or porous material, moisture movement is mainly by vapour diffusion through the void or empty spaces. D_{eff} and velocity of moisture movement within the material are relatively related while drying rate is the rate at which moisture vaporizes to the surrounding air or a change of moisture to vapour by evaporation which depends largely on the pressure difference existing between the food material and surrounding air as a result of temperature difference (So'bah et al., 2017).

Pre-drying treatment of fruits and vegetables has been known to favour or disfavour their drying rates which is a function of both internal mass transport mechanisms and external heat (Mauro et al., 2005). It helps retain food sensory and nutritional qualities; as previous researches have shown that the effects of drying-air conditions, most especially drying-air temperature have adversely affected the quality attributes of tomato if not properly controlled.

Therefore, it becomes imperative to investigate the effects of pre-drying treatment and drying-air temperature on moisture ratio and effective moisture diffusivity of tomato.

2. MATERIALS AND METHODS

2.1 Raw Material

Fresh tomato samples of three different varieties were obtained from local growers in Oteh area, a surburb of Ilorin Kwara State of Nigeria. The samples were sorted visually according to their ripeness, firmness and size. Samples were thoroughly washed under tap water, sliced using a stainless steel knife, deseeded with a needle and blanched in hot water for 1 min at 90°C to minimize browning and enzymatic reaction during drying process.

2.2 Pre-drying treatment Process

Sliced samples were divided into five parts and were subjected to further pre-drying treatments following the mechanical and thermal pre-drying treatments used on them initially. These other methods include treatment in osmotic and chemical solutions. The first, second, third and fourth parts were treated in a mixture of honey and sugar solution at 40°Bx concentration (honey: sugar: water ratio 2: 1: 1.9) for 10 min which was prepared using a refractometer (Model: M10481, by ABBE MARK II, USA), 2% ethyl acetate solution for 1 min, 0.5g/100ml NaCl solution for 10 min and 1% $MgCl_2 \cdot 6H_2O$ and 0.5% $Na_2S_2O_5$ for 10 min respectively. And the fifth part served as the control sample which was immersed in distilled water at room temperature for 10 min. Each sample weighed 250 g with an electronic balance (Model: WH-B06, sensitivity ± 0.01g by WEIHENG, China) before pre-drying treatment. After pre-drying treatment samples were drained and bloated with absorbent paper.

2.3 Drying Procedure

After pre-drying treatments of the samples they were dried in an automated forced convection cabinet dryer (FCCD) designed for the purpose of this research at different drying-air temperatures (45, 55 and 65°C), the dryer was run to attain the desired temperature before loading the samples on its labelled

trays. The trays were rested on load cells which were linked to a microcontroller which was in turn connected to a computer system which has a software with Arduino programme used in monitoring and controlling the dryer. The dryer was also equipped with a thermo-hygrometer sensor used in sensing the drying-air temperature and humidity and three (3) solid state relays which were used for switching on and off the two (2) heaters (3.6 kW) and a centrifugal fan (2m/s). The dryer was pre-selected to take record of every 5 min as the drying experiment progressed. The measurements recorded were the instantaneous weight on each of the four (4) trays, the drying-air temperature and humidity. This dryer totally eliminates the drudgery, time and energy wastages associated with previous drying experiments when samples were brought out to measure. The FCCD is shown in Figure 1.
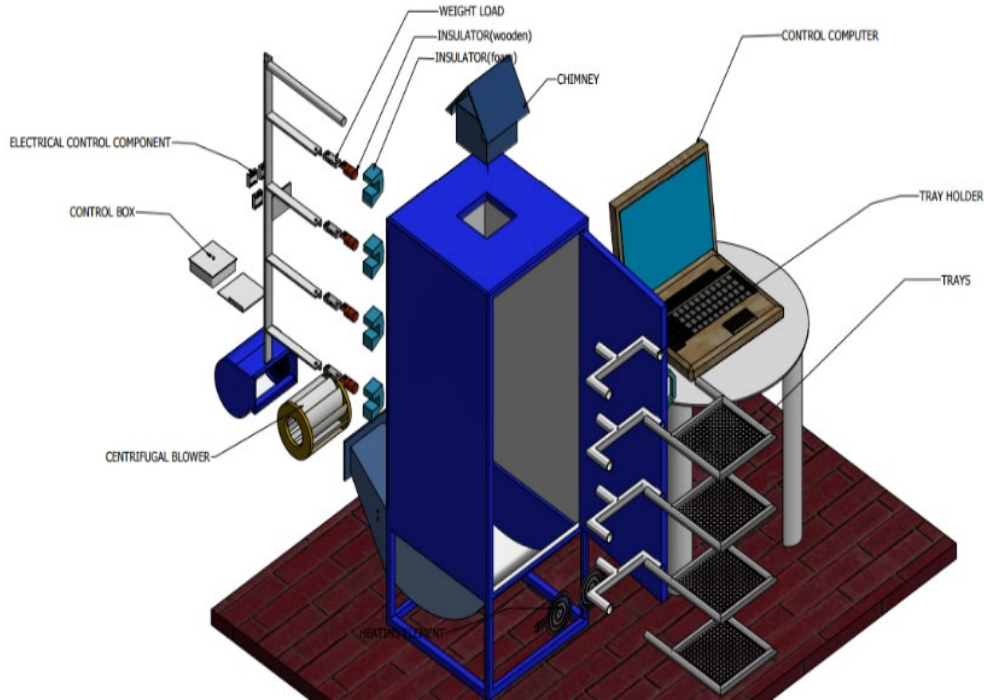
Figure 1. 3D View of the Automated FCCD

2.4   Determination of output Parameters

2.4.1 Instantaneous Moisture Content (M_t)

The instantaneous moisture content (M_t) of tomato at any given time (t) during the drying experiment was estimated using Equation 1;

$$M_t = \frac{(M_o + 1) - W_t - 1}{W_o} \tag{1}$$

where;
M_t = Instantaneous m.c. (% wb)
M_o = Initial m. c. (% wb)
W_t = Weight of product at any time, t during drying (g)
W_o = Initial weight of the sample (g)

2.4.2 Moisture Ratio (MR)

Moisture ratio was calculated as expressed in Equation 2;

$$MR = \frac{M_t - M_e}{M_o - M_e} \tag{2}$$

where;
M_t = m.c. of the tomato samples at any time t (% db)
M_e = Equilibrum m.c. of the tomato samples (% db)
M_o = Initial m.c. of the samples before drying (% db)

2.4.3 Effective Moisture Diffusivity

The effective moisture diffusivity (D_{eff}) was estimated using the "simplified mathematical Fick's second diffusion model". The solution of Fick's second law in slab geometry, having the following assumptions; that moisture migration is strictly dependent on diffusion, shrinkage is negligible, diffusion coefficients are constant and temperature which was the diffusion model was simplified to linear equation by Crank (1975) as it is expressed in Equation 3.

$$MR = \frac{M}{M_O} = \frac{8}{\pi^2} \sum_{n-1}^{\infty} \frac{1}{(2n-1)^2} \exp\left[\frac{(2n-1)^2 \pi^2 D_t}{4L^2}\right]$$ (3)

where,
MR = Moisture Ratio
M = Moisture content at any time (kg water/kg dry matter)
M_o = Initial moisture content (kg water/kg dry matter)
n = 1,2,3,... the number of terms taken into consideration
t = time of drying in seconds
D_t = Effective moisture diffusivity (m²/s)
L = thickness of the slice (m)

Equation 4 was used since the drying process involved a long term drying due to high moisture content present in tomato

$$MR = \frac{8}{\pi^2} \exp\left[\frac{\pi^2 D_t}{4L^2}\right]$$ (4)

The slope (K_o) of the graph was estimated by plotting *ln (*MR) against time (t) as presented in Equation 5;

$$K_o = \frac{\pi^2 D}{4L^2}$$ (5)

2.5 Design of Experiment

The experiment was designed using the Box-behnken design (BBD) of response surface framework of Design Expert Software V 6.0.6 (US, Stat-Ease Inc.) resulting in 68 runs. The experimental input parameters were sample variety (*Hausa, Tiwantiwa* and Roma *VFN*), slice thickness (5.0 mm, 7.5 mm and 10 mm), pre-drying treatment (Ethyl acetate, $MgCl_2 \cdot 6H_2O$ and $Na_2S_2O_5$, NaCl and Honey and Sugar) and drying-air temperature (45, 55 and 65°C).

2.56 Statistical Analysis
In this experiment statistical analysis of responses were done using quadratic model interface of the Design Expert software with alpha to exit 0.050 and regression coefficients were obtained.

3. RESULTS AND DISCUSSION

3.1 Effects of Input Parameters on Moisture Ratio and Effective Moisture Diffusivity

3.1.1 Effect of Drying-air Temperature, Slice Thickness, Variety and Pre-drying Treatments on Moisture Ratio

The effect of drying-air temperature on the moisture ratio (MR) of tomato samples is shown in Figure 2 a, it shows that samples subjected to 65°C had average MR of 70.19%, 55°C had 69.13%, while those subjected to 45°C had the highest average MR of 77.54%. This result agrees with the findings of Yousefi et al. (2013) on the drying of papaya slices at 40, 50 and 60°C. The highest MR in samples dried at 45°C can be attributed to the slowest rate at which
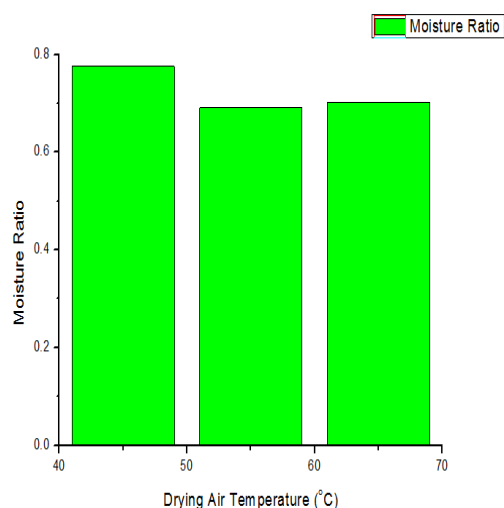
Figure 2 a: Effect of Air-temp. on Moisture Ratio
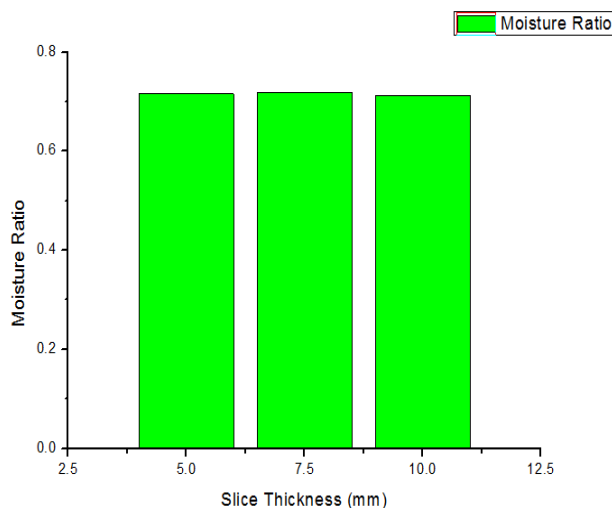
Figure 2 b: Effect of Slice Thickness on Moisture Ratio
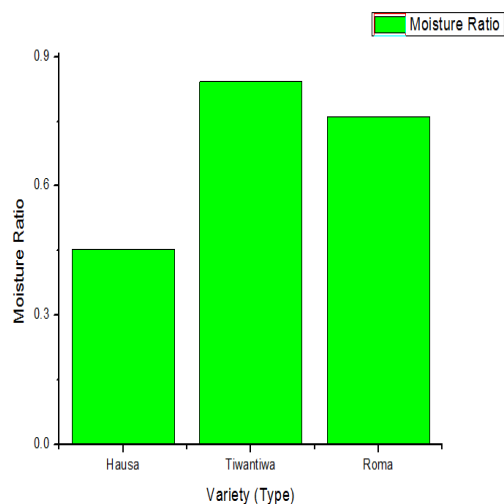
Figure 2 c: Effect of Variety on Moisture Ratio
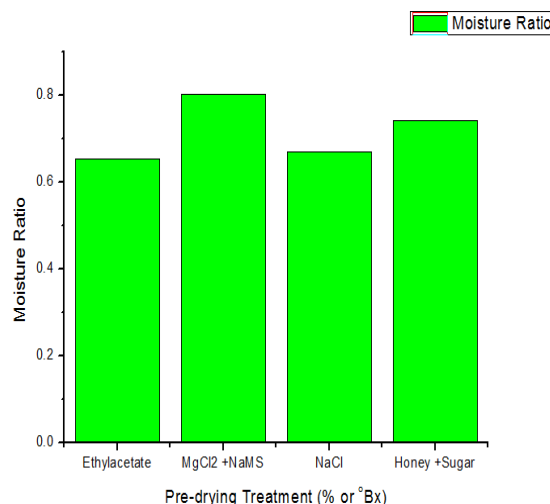
Figure 2 d: Effect of Pre-drying Treatment on Moisture Ratio

Table 1: Analysis of Variance for Moisture Ratio of Tomato Slices

Source	Sum of Squares	DF	Mean Square	F Value	Prob > F	
Model	2.32	21	0.11	11.95	< 0.0001	*Sig.*
A	0.061	1	0.061	6.60	0.0137	
B	2.979E-005	1	2.979E-005	3.217E-003	0.9550	
C	0.90	1	0.90	97.05	< 0.0001	
D	0.21	3	0.071	7.63	0.0003	
A^2	9.739E-003	1	9.739E-003	1.05	0.3107	
B^2	2.642E-003	1	2.642E-003	0.29	0.5959	
C^2	0.93	1	0.93	100.38	< 0.0001	
AB	3.169E-004	1	3.169E-004	0.034	0.8541	
AC	0.034	1	0.034	3.69	0.0611	
AD	0.057	3	0.019	2.07	0.1184	
BC	5.621E-003	1	5.621E-003	0.61	0.4401	
BD	6.219E-003	3	2.073E-003	0.22	0.8793	
CD	0.11	3	0.035	3.83	0.0160	
Residual	0.41	44	9.260E-003			
LOF	0.40	28	0.014	25.55	0.0001	
Pure Error	8.914E-003	16	5.571E-004			
Cor Total	2.73	65				

LOF-Lack of Fit; Significance Level (p≥0.05)

moisture was diffusing out of the samples in which more moisture was retained in the samples compared with those dried at 55°C and 65°C. Moisture gets more excited at higher drying-air temperature and diffuses. ANOVA Table 1, revealed that the effect of drying-air temperature (D) was significant on the MR of samples at significance level of $p \leq 0.05$.

The effect of slice thickness on MR is shown in Figure 2 b. It is seen that the MR of samples sliced at 5.0 mm, 7.5 mm and 10 mm were 71.59 %, 71.89% and 71.29% respectively. This values were seen to be very close and the ANOVA Table 2 shows that the effect of slice thickness (B) was not statistically significant ($p \leq 0.05$) on the MR.

The effect of samples' variety on the MR of tomato is shown in Figure 2 c, it was found that *Hausa* variety had an average MR of 45.27%, *Tiwantiwa* variety had 84.25% and Roma *VFN* variety had 76.03%, these show that *Tiwantiwa* variety has the highest MR and *Hausa* variety has the lowest. This can be attributed to the high initial m.c. of *Tiwantiwa* variety which was 95.82% and low moisture content of *Hausa* variety which was 85.03%. While that of Roma *VFN* variety had initial moisture content of 93.76%. The difference in MR of the samples might be attributed to the microstructural characteristics of the tomato varieties as the cells of *Tiwantiwa* variety might be less porous and able to retain more moisture compared with others. The ANOVA Table 1 further reveals it that statistically the individual effect of tomato variety (C), its quadratic effect (C^2) and the interactive effect CD between variety and pre-drying treatment were highly significant statistically at $p \leq 0.05$ on the MR.

The effect of samples' pre-drying treatment on MR of treated tomato samples is shown in Figure 2 d samples pre-treated with ethyl acetate solution had 65.34% MR, those with $MgCl_2 \cdot 6H_2O$ and $Na_2S_2O_5$ solution had 80.21%, while those with NaCl solution had 66.95%, those pre-treated with mixture of honey and sugar solution had 74.11% and control samples had an average MR of 74.09%. This

observation would result from the hydrophilic property of $MgCl_2 \cdot 6H_2O$ which makes it highly hygroscopic in nature and therefore samples pre-treated in it has the highest MR. Ethyl acetate easily vaporizes into the air and not hygroscopic in nature and therefore was seen to have the least MR among others. As further shown by the ANOVA Table 1 statistically the effect of pre-drying treatment was significant on the MR of treated samples ($p \leq 0.05$).

3.1.2 Effects of Drying-air Temperature, Slice Thickness, Variety and Pre-drying Treatments on Effective Moisture Diffusivity

The effect of drying-air temperature on the effective moisture diffusivity (D_{eff}) of tomato samples is shown in Figure 3 a, it was seen that samples subjected to 65°C had highest D_{eff} of 1.69 x 10^{-8} m^2/s, 55°C had 1.20 x 10^{-8} m^2/s, while those subjected to 45°C had the lowest D_{eff} of 5.99 x 10^{-9} m^2/s, this agrees with the findings of Yilmaz et al. (2017), that increase in air temperature led to increase in D_{eff}. Results with this trend had been reported earlier by Jaiyeoba and Raji (2012) who had worked on the estimation of D_{eff} of Tomato and found that D_{eff} increases with increase in air temperature and also found that the D_{eff} of tomato was within 10^{-8} m^2/s. The highest D_{eff} observed in samples dried at 65°C can be attributed to the high level of drying-air temperature used which contains more heat energy required to activate the movement of water from the internal part of the products to their surface for drying to occur. As reported by Mewa et al. (2018), that water activity increases with increase in temperature which results in increase in D_{eff} of beef during drying. Analysis of Variance (ANOVA) Table 2, shows that the effect of drying-air temperature was highly significant on the D_{eff} of samples at $p \leq 0.05$, therefore the findings of Yilmaz et al. (2017) was replicated that effect of drying-air temperature was significant on the D_{eff} of pomegranate fruit leather.
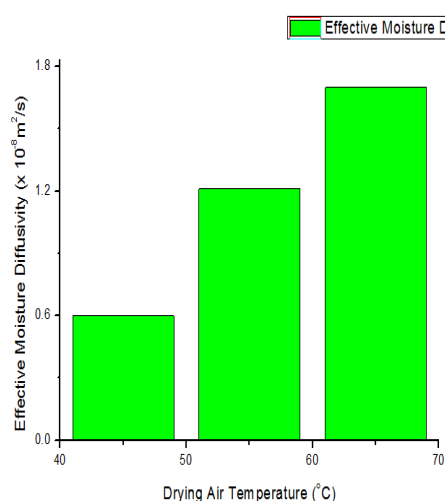
Figure 3 a: Effect of Air-temp. on Effective Moisture Diffusivity
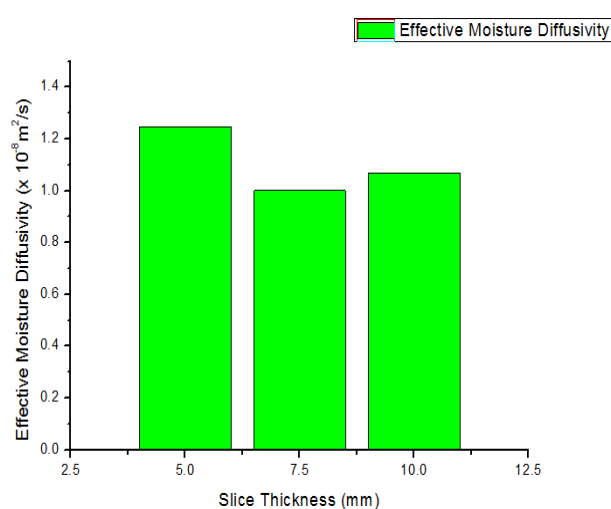
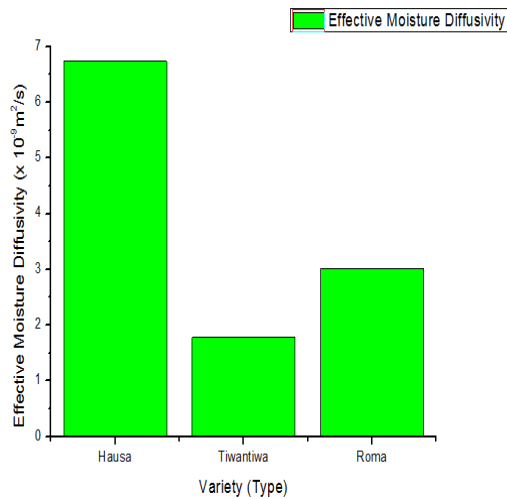Figure 3 b: Effect of Slice Thickness on Eff. Moisture Diffusivity

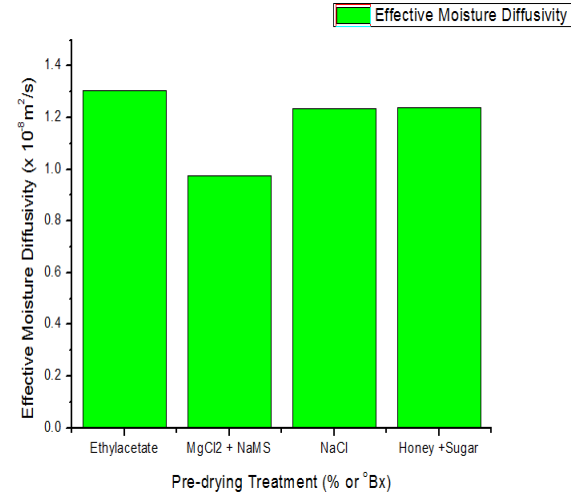Figure 3 c: Effect of Variety on Eff. Moisture Diffusivity

Figure 3 d: Effect of Pre-drying Treatment on Eff. Moisture Diffusivity

Table 2: Analysis of Variance for Effective Moisture Diffusivity of Tomato Slices

Source	Sum of Squares	DF	Mean Square	F Value	Prob.>F	
Model	4.107E-015	21	1.956E-016	4.81	< 0.0001	*Sig.*
A	1.043E-015	1	1.043E-015	25.63	< 0.0001	
B	1.575E-017	1	1.575E-017	0.39	0.5369	
C	1.574E-016	1	1.574E-016	4.36	0.0408	
D	1.040E-016	3	3.465E-017	0.85	0.4728	
A^2	9.811E-018	1	9.811E-018	0.24	0.6257	
B^2	1.433E-017	1	1.433E-017	0.35	0.5557	
C^2	2.492E-015	1	2.492E-015	61.26	< 0.0001	
AB	1.699E-019	1	1.699E-019	4.177E-003	0.9488	
AC	6.786E-019	1	6.786E-019	0.017	0.8978	
AD	2.452E-016	3	8.175E-017	2.01	0.1257	
BC	6.958E-020	1	6.958E-020	1.711E-003	0.9672	
BD	2.275E-017	3	7.582E-018	0.19	0.9051	
CD	2.528E-017	3	8.428E-018	0.21	0.8909	
Residual	1.871E-015	46	4.068E-017			
LOF	1.231E-015	30	4.103E-017	1.03	0.4949	
Pure Error	6.400E-016	16	4.000E-017			
Cor Total	5.978E-015	67				

LOF-Lack of Fit; Significance level (p≥0.05)

Effect of slice thickness on D_{eff} is found in Figure 3 b. The D_{eff} of tomato samples sliced at 5 mm, 7.5 mm and 10 mm were 1.25, 1.00 and 1.07 $x10^{-8}$ m^2/s respectively. These values were seen to be quite close and the ANOVA Table 2 shows that the effect of slice thickness was not significant statistically at p ≤ 0.05 on the D_{eff}. However, this is not in agreement to the report by Yilmaz et al. (2017) that increase in slice thickness results in increase in effective moisture diffusivity and states that slice thickness was statically significant on D_{eff}.

The effect of samples' variety on the D_{eff} of tomato is shown in Figure 3 c, it was found that *Hausa* variety had an average D_{eff} of 6.73 $x10^{-9}$ m^2/s, Tiwantiwa variety had 1.78 $x10^{-9}$ m^2/s and Roma *VFN*

variety had 3.02 x10^{-9} m^2/s, these show that *Hausa* variety had the highest D$_{eff}$ and *Tiwantiwa* variety had the lowest. These show that the samples with the highest moisture content tend to have lower effective moisture diffusivity than those with the lowest moisture content which have higher effective moisture diffusivity. *Tiwantiwa* variety had the highest initial moisture content of 95.82% and MR while *Hausa* variety had lowest initial moisture content of 85.03% and lowest MR. Roma *VFN* variety has initial moisture content of 93.76% and average MR. This was in agreement with the findings Sharma and Prasad (2004) in which it was found that D$_{eff}$is a function of samples' m.c. which increases gradually with decrease in m.c. The reason for this was that as m.c. decreased vapour phase diffusivity increased provided the pores were kept opened. The ANOVA Table 2 further reveals it that statically the individual effect of tomato variety (C) and its quadratic effect (C^2) were highly significant (p ≤ 0.05) on the D$_{eff}$.

The effect of samples' pre-drying treatment on D$_{eff}$. of treated tomato samples is shown in Figure 3 d, samples pre-treated with ethyl acetate solution had 1.31 x10^{-8} m^2/s D$_{eff}$, those with MgCl$_2$·6H$_2$O and Na$_2$S$_2$O$_5$ solution had 9.76 x10^{-9} m^2/s, while those with NaCl solution had1.23 x10^{-8} m^2/s, those pre-treated with mixture of honey and sugar solution had1.24 x10^{-8} m^2/s and control had 8.425 x 10^{-9} m^2/s. Samples pre-treated with ethyl acetate solution had the highest effective moisture diffusivity while those pre-treated in MgCl$_2$·6H$_2$O and Na$_2$S$_2$O$_5$ solution had the lowest. From Figure 2 d it can be seen that samples pre-treated in ethyl acetate had the lowest MR while those pre-treated in MgCl$_2$·6H$_2$O and Na$_2$S$_2$O$_5$ solution had the highest MR. Therefore, the higher the MR the lower the effective moisture diffusivity and vice versa. This claim had also been found by Sharma and Prasad (2004) and Darvishi et al. (2016). As further shown by the ANOVA Table 2 statistically the effect of samples' pre-drying treatment was not significant on the D$_{eff}$ of treated samples (p ≤ 0.05).

4. CONCLUSION

1. Moisture ratio of tomato at any stage during the drying process reduces with increase in drying-air temperature but effective moisture diffusivity increases with increase in drying-air temperature

2. The moisture ratio of tomato was dependent on its variety which can be linked to its initial moisture content, also Pre-drying treatment of samples had strong influence on samples' MR and effective moisture diffusivity which can either increase or decrease it.

3. Samples' lowest MR were obtained under these conditions; processed *Hausa* variety, 10 mm thickness, Ethyl acetate pre-drying treatment and dried at 55°C while highest effective moisture diffusivity was obtained under these conditions processed *Hausa* variety, 5 mm thickness, Honey and Sugar pre-drying treatment and dried at 65°C in a FCCD.

4. Processed *Hausa* variety is more preferable to other varieties of tomato used; as it promotes low MR and high effective moisture diffusivity during the drying process. This will help reduced energy consumed during drying of tomato and will save processing time.

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Oral Session | Postharvest/Food Technology and Process Engineering

[5-1015-C] Postharvest/Food Technology and Process Engineering (5)

Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[5-1015-C-04] Extending the Shelf-life of Upland Water Spinach (*Ipomoea aquatica*) Using Trimming, Modified Atmosphere Packaging (MAP) and Low-Temperature Storage

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Keywords: upland water spinach, modified atmosphere packaging, low temperature storage, postharvest, leafy vegetable

Upland Water Spinach (UWS) is highly perishable in nature and have a short shelf life (2 days). Small-scale farmers, traders, and restaurant owners sought simple, low-cost techniques to prolong its shelf-life. In this study, the effects of trimming of roots, modified atmosphere packaging (MAP) using polyethylene bag with 1 pinprick, and low temperature storage (20 ± 0.5°C and 10 ± 0.5ºC) on the shelf life of UWS were evaluated based on its visual quality, yellowing, wilting, disease incidence and shelf-life. Results showed that at room temperature storage (29 ± 1.0ºC), packed UWS had a higher shelf life (3days) compared to the unpacked ones (2 days), regardless of the presence of roots. At 20 ± 0.5°C storage, unpacked UWS without roots had a longer shelf life (3days) than those with roots (2days). Packed UWS at 20 ± 0.5ºC, regardless of the presence of roots, had a longer shelf life (4 days) as compared to the unpacked ones. At 10 ± 0.5ºC storage, unpacked UWS had a shelf life of 3 days. Among all treatments, packing UWS without roots in PEB with 1 pinprick in combination with storage at 10 ± 0.5ºC extends the shelf life to 5 days, with notable delay in occurrence and reduction of the extent of wilting and yellowing. This practice can be used by small-scale farmers, traders, and restaurant owners to reduce daily procurement costs incurred for transportation, hauling and manpower.

Extending the Shelf-life of Upland Water Spinach (*Ipomoea aquatica*) Using Trimming, Modified Atmosphere Packaging (MAP) and Low-Temperature Storage

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ABSTRACT

Upland Water Spinach (UWS) is highly perishable in nature and have a short shelf life (2 days). Small-scale farmers, traders, and restaurant owners sought simple, low-cost techniques to prolong its shelf-life. In this study, the effects of trimming of roots, modified atmosphere packaging (MAP) using polyethylene bag with 1 pinprick, and low temperature storage (20 ± 0.5°C and 10 ± 0.5°C) on the shelf life of UWS were evaluated based on its visual quality, yellowing, wilting, disease incidence and shelf-life. Results showed that at room temperature storage (29 ± 1.0ºC), packed UWS had a higher shelf life (3days) compared to the unpacked ones (2 days), regardless of the presence of roots. At 20 ± 0.5°C storage, unpacked UWS without roots had a longer shelf life (3days) than those with roots (2days). Packed UWS at 20 ± 0.5ºC, regardless of the presence of roots, had a longer shelf life (4 days) as compared to the unpacked ones. At 10 ± 0.5ºC storage, unpacked UWS had a shelf life of 3 days. Among all treatments, packing UWS without roots in PEB with 1 pinprick in combination with storage at 10 ± 0.5ºC extends the shelf life to 5 days, with notable delay in occurrence and reduction of the extent of wilting and yellowing. This practice can be used by small-scale farmers, traders, and restaurant owners to reduce daily procurement costs incurred for transportation, hauling and manpower.

Keywords:
Upland water spinach
Modified atmosphere packaging
Low temperature storage
Postharvest
Kangkong

1.   INTRODUCTION

Upland Water Spinach, also known as upland *kangkong* in the Philippines, is a leafy vegetable that grows rapidly (~25-30 days), is easily cultivated, and can thrive in most soil types (Goebel *et al.,* 2010) and various seasons throughout the year (Science and Development Network, 2013). In developing countries like Philippines, the importance of UWS has been recognized due to its availability in the market at a remarkably low price (Prasad *et al.,* 2008). Young leaves, petioles and stems used as viand, cooked alone or with meat or fish, while vines are used as fodder for cattle and pigs. More attention is continuously drawn to UWS due to its high nutritional value (protein, fiber, calcium, magnesium, iron, vitamins A, C, and E, folic acid, and phenolic compounds) and better appearance than low land water spinach (Dua *et al.,* 2015), as both market and consumers are driven towards healthier food choices.

UWS is highly perishable in nature and have a short shelf life (2 days). Effective yet simple and low-cost techniques in prolonging the shelf-life of UWS is sought by farmers, traders, as well as restaurants. Farmers and traders believe that trimming the roots of UWS will hasten its deterioration hence, selling UWS with roots has been their common practice. However, the roots can add to the bulk of vegetable handled and transported, may be source of contaminants in which concerns on safety and quality may arise, and may cause fraudulent addition to weight and price that can be burdensome for customers. This theory in trimming of roots was tested in this study.

Various techniques on modified atmosphere packaging (MAP), appropriate storage temperatures, and minimal processing were explored to maintain quality and reduce losses in leafy vegetables as recommended in published literatures such as Kitinoja and Kader (2002), Cantwell and Suslow (2006), Kanlarayat (2007), and Acedo (2010). However, the use of these techniques has not been reported for UWS in the Philippines. Hence, these techniques were studied to match the needs of concerned UWS small-scale farmers, traders and restaurant owners.

This study aims to prolong the shelf-life of upland water spinach using simple and low-cost techniques by determining the effect of trimming the roots, MAP using polyethylene bags with pinprick, and low temperature storage (20±0.5°C and 10±0.5ºC) on the shelf-life of UWS.

2. MATERIALS AND METHODS
2.1. Plant Materials
Freshly harvested 30 days old UWS with roots were obtained from a nearby vegetable farm in UPLB. These were placed in clean 20-kg capacity plastic crates and hauled immediately to the Postharvest Horticulture Training and Research Center (PHTRC) laboratory.
2.2. Sample Preparation
Damage-free UWS plants with tender leaves and stems were selected from the harvest pool. In the packing house, these were washed thoroughly using tap water and sanitized using 100 ppm hypochlorite solution (Suslow, 2000) and then drained. Samples were then air-dried in the minimal processing laboratory (operating temperature: 25ºC).
2.3. Treatment
Completely dried UWS were distributed for each treatment as stipulated in Table 1. Packed samples were tape-sealed in 0.02mm polyethylene bag (PEB) with 1 pinprick. Treatments were selected based on best practices taken from preliminary studies.

Table 1. Postharvest techniques to prolong the shelf-life of UWS.

Treatment No.	Packaging PEB / UNP[1]	Presence of Roots WR / NR[2]	Storage Temperature ºC
1	PEB	WR	29±1.0ºC
2	PEB	NR	29±1.0ºC
3	UNP	WR	29±1.0ºC
4	UNP	NR	29±1.0ºC
5	PEB	WR	20±0.5ºC
6	PEB	NR	20±0.5ºC
7	UNP	WR	20±0.5ºC
8	UNP	NR	20±0.5ºC
9	PEB	NR	10±0.5ºC

| 10 | UNP | NR | 10±0.5ºC |

[1] packed in polyethylene bag with one pinprick = PEB, unpacked = UNP;
[2] with roots = WR; without / no roots = NR; N=100

Permeating the package with a pinprick-sized air passage creates a naturally induced modified atmosphere for the product, providing protection for water loss while allowing enough respiration to occur through the hole. Modified atmosphere packaging (MAP) is retains freshness and extends shelf life of fresh produce by inhibiting moisture loss and slow down respiration thereby maintaining its color, reducing loss due to product respiratory heat, and maintaining the natural fresh taste of produce (Acedo, 2010).

Unpacked samples were bundled using rubber bands and placed uncovered on clean trays, simulating storage practices of UWS in Filipino households. Packed and unpacked UWS were placed in temperature simulations of room temperature (29±1.0ºC), open-type display chiller temperature (20±0.5ºC) and door-type display chiller temperature (10±0.5ºC). These storage temperatures were chosen as these are used in local restaurants that sell fresh UWS and offer UWS in their menu. Each replicate weighs 250±0.50 grams and there are 10 replicates for each temperature studied.

2.4.   Data Collection and Analysis

Being a leafy vegetable, the quality of upland water spinach is mainly based on appearance that can be discerned by the human senses such as freshness, shape, size, maturity, color, turgidity, freedom from defects such as rot, physical damage, yellowing, or wilting (Acedo, 2010). In this study, these parameters were scored using Visual Quality Rating (VQR) to consider all visual factors that may affect the physical appearance of commodities. Shelf-life was determined by the number of days wherein the samples are edible.

Visual quality, yellowing, wilting and disease incidence were evaluated daily using indices developed by PHTRC-UPLB (Table 2, 3). Samples were evaluated daily for these parameters until it surpassed the limit of marketability (VQR=3). All data obtained were subjected to statistical analyses using SAS V9.0.

Table 2. Visual quality rating for fruits and vegetables (Horticulture 109.1 Laboratory Manual, PHTRC-UPLB).

Visual Quality Rating	Description
9,8	Excellent, field fresh
7,6	Very good, trace defects
5,4	Good, defects minor
3	Fair, defects moderate, limit of marketability
2	Poor, defects serious, limit of edibility
1	Non-edible under usual condition

Table 3. Indices for wilting, yellowing, and disease incidence (Horticulture 109.1 Laboratory Manual, PHTRC-UPLB).

Index	Description		
	Wilting	Yellowing	Disease Incidence
1	None	Absent	Absent
2	Trace, <10% leaves wilted (mostly tips and edges)	Slight (up to 20% leaves discolored)	Slight (up to 20% leaves infected)
3	Slight, 10-25% leaves wilted	Moderate (21-40% leaves discolored)	Moderate (21-40% leaves infected)

4	Moderate, 26-50% leaves wilted	Severe (>40% leaves discolored)	Severe (>40% leaves infected)
5	Severe, <50% leaves wilted	Not applicable	Not applicable

3.   RESULTS AND DISCUSSION

3.1.   Visual Quality Rating (VQR) and Shelf-life

VQR for UWS were shown in Figure 1.a-c. Regardless of the type of packaging, trimming of roots did not affect VQR at 29±1.0ºC. PEB-packed UWS remained marketable until day 2, which shows that packaging in PEB with 1 pinprick extends the shelf life by 1 day at 29±1.0ºC (Table 4). On the other hand, rapid deterioration on visual quality can be observed in unpacked UWS at 29±1.0ºC from day 1 to day 2.

In samples stored at 20±0.5ºC, PEB-packed UWS were marketable until the fourth day of storage, regardless of the presence of roots while unpacked samples with roots had increased its shelf-life by 1 day. It can be noted that the shelf-life of PEB-packed samples, regardless of the presence of roots, also increased by one day at 20±0.5ºC.

At 10±0.5ºC, PEB-packed samples were marketable up to 5 days of storage while unpacked samples lasted for 3 days. The rapid decline in VQR of unpacked samples on the second storage day is notable.

The presence of roots did not have effects on the VQR and shelf-life of packed samples (Figure 1.a and b). This can be attributed to the protection provided by the packaging which inhibits water loss thereby retaining freshness of the leaves (Acedo, 2010). However, retails packs and bundles with trimmed roots are fuller in terms of useful portions (young leaves and stems) which may provide more value for the price of each pack and benefit consumers.

With the base UWS shelf-life of 2 days (control), storage in 20±0.5ºC and 10±0.5ºC in conjunction with PEB-packing with one pinprick increased the shelf-life of UWS by 1 day and 2 days, respectively. Among all samples in different storage temperatures, PEB-packed UWS without roots and stored at 10±0.5ºC had the superior VQR and remained marketable up to 5 days of storage.
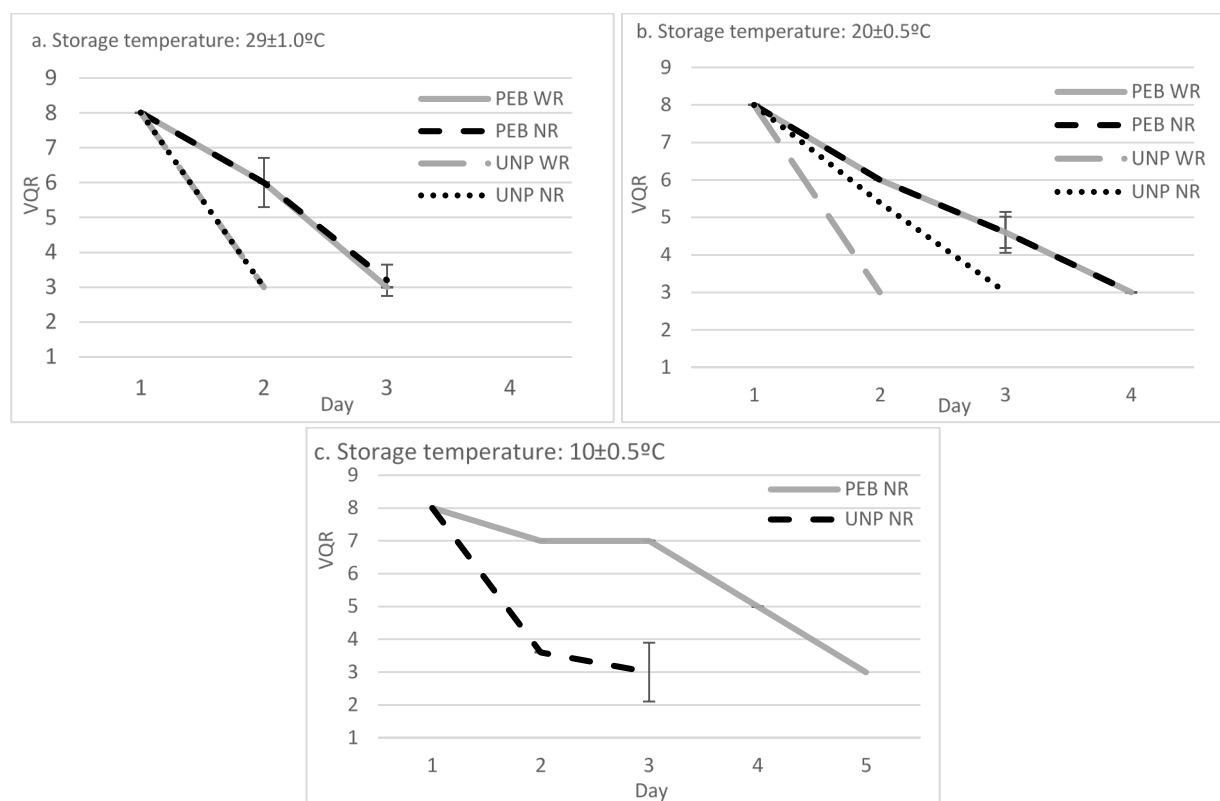
Figure 1.a-c. Visual quality rating for UWS stored at 29±1.0ºC, 20±0.5ºC and 10±0.5ºC (Packed in polyethylene bag with one pinprick = PEB, unpacked = UNP; with roots = WR; without / no roots = NR; VQR scores: 9,8= Excellent, field fresh, 7,6= Good, defects minor, 5,4= Fair, defects moderate, limit of marketability; 3= Poor, defects serious, 2=Limit of edibility, 1=non-edible under usual condition; N=100).

Table 4. Shelf-life of UWS stored at 29±1.0ºC, 20±0.5ºC and 10±0.5ºC ([1] Packed in polyethylene bag with one pinprick = PEB, unpacked = UNP; [2] with roots = WR; without / no roots = NR; N=100).

Treatment	Packaging	Presence of Roots	Storage Temperature	Shelf-life [3]
No.	PEB / UNP[a]	WR / NR[b]	ºC	Days
1	PEB	WR	29±1.0ºC	3c
2	PEB	NR	29±1.0ºC	3c
3	UNP	WR	29±1.0ºC	2d
4	UNP	NR	29±1.0ºC	2d
5	PEB	WR	20±0.5ºC	4b
6	PEB	NR	20±0.5ºC	4b
7	UNP	WR	20±0.5ºC	2d
8	UNP	NR	20±0.5ºC	3c
9	PEB	NR	10±0.5ºC	5a
10	UNP	NR	10±0.5ºC	3c

[3] *Shelf-life values followed by similar letters are <u>not</u> significantly different from each other.*

3.2.   Yellowing

Yellowing was observed on all samples at the second day of storage except for the PEB with roots at 20±0.5ºC and samples at 10±0.5ºC (Figure 2.a-c.). Packed samples exhibited discoloration which can be attributed to exposure to its own ethylene. While MAP slows down respiration and protects the leaves from moisture loss, ethylene produced during senescence can build up inside the packaging with prolonged storage. This may cause yellowing, epinasty (leaf curving) and abscission (Cantwell and Suslow, 2006).

At 10±0.5ºC, yellowing was delayed until the third day of storage. This can be attributed to the effect of cold storage which slows down the rates of physiological changes that the commodity undergoes, thereby reducing its effects such as discoloration (Kitinoja and Kader, 2002).

No significant trends were observed on the response of UWS to MAP and presence of roots in relation to yellowing.
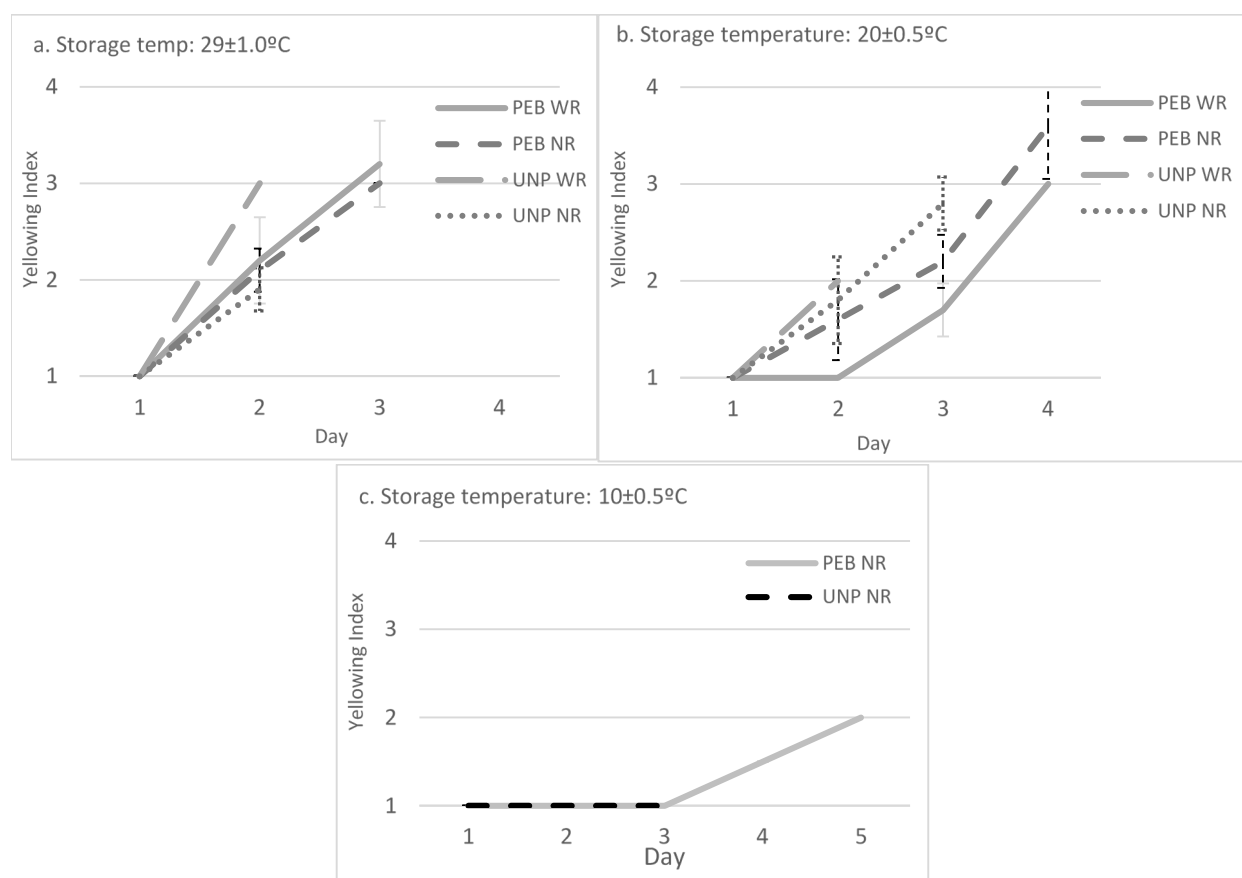
Figure 2.a-c. Yellowing in UWS stored at 29±1.0ºC, 20±0.5ºC and 10±0.5ºC (Packed in polyethylene bag with one pinprick = PEB, unpacked = UNP; with roots = WR; without / no roots = NR; Yellowing Index:1=absent, 2=Slight or 20%, 3= Moderate or 21-40%, 4=Severe or >40% of leaves discolored; N=100).

3.3. Wilting

Unpacked UWS had a significantly higher rate of wilting compared to the PEB-packed with 1 pinprick UWS in all storage temperatures (Figure 3.a-c.). This exhibits the effectiveness of MAP to decrease rates of moisture loss (Kitinoja and Kader, 2003).

Wilting was observed in PEB-packed samples only on the third day of storage in 29±1.0ºC and 20±0.5ºC. For the PEB-packed UWS in 10±0.5ºC, the incidence of wilting was delayed up to 4 days. On the other hand, no significant trends were observed on the response of UWS to presence of roots in relation to wilting.

It is important to reduce the incidence of wilting because it promotes degradation of nutritional components (e.g. vitamins and minerals) and imposes stress (i.e. water stress) that increases respiration and ethylene production. This should be prevented to maximize the health benefits of the vegetable. According to Kanlayanarat (2007), 5-10% in fresh weight make leafy vegetables appear wilted and unusable.
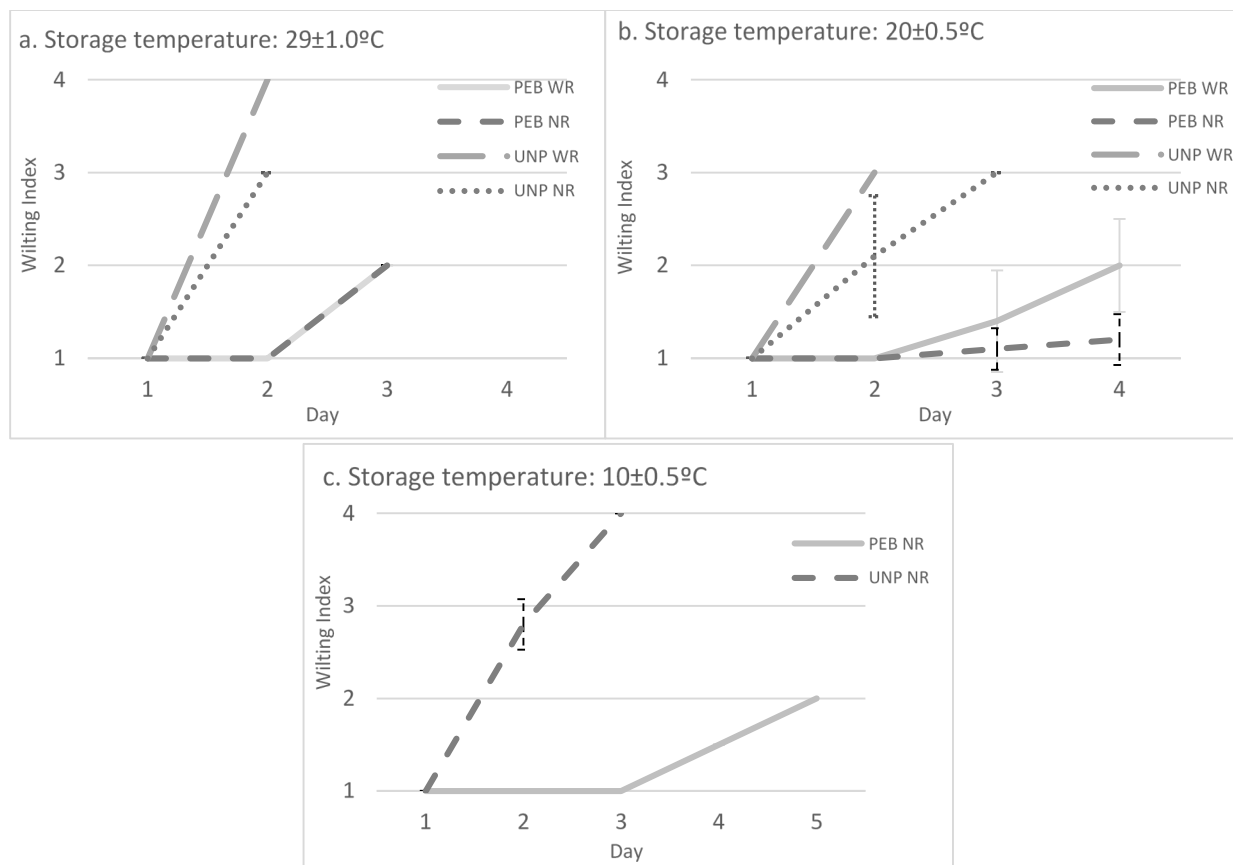
Figure 3.a-c. Wilting in UWS stored at 29±1.0ºC, 20±0.5ºC and 10±0.5ºC (Packed in polyethylene bag with one pinprick = PEB, unpacked = UNP; with roots = WR; without / no roots = NR; Wilting Index:1=none, 2=trace or <10%, 3=slight or 10-25%, 4=moderate or 25-50%, 5=severe or >50% wilted leaves; N=100).

3.4.   Disease Incidence

Disease incidence was not observed on all samples (data not shown). This can be attributed to the systematic preparation of samples including washing with 100ppm hypochlorite solution and proper air drying.

According to the Philippine National Standards (Bureau of Agriculture and Fisheries Standards, 2016), washing with sodium hypochlorite solution is allowable for organically produced vegetables. The most effective concentration at the safe range is 100ppm (Suslow, 2000). Application of sanitizing agents may

help minimize the risk of a variety of biological hazards or contaminants such as *Salmonella sp., Escherichia coli, Listeria sp.,* and mycotoxins that pose food safety concerns leading to outbreaks (Herman *et al.,* 2015). At the same time, sanitation practices also reduce risk for bacterial soft rot, commonly caused by *Erwinia carotovora* (Tournas, 2005)*.* This causes decay especially in packed leafy vegetables, since the packaging promotes build up of moisture that is promotes bacterial growth.

4.   CONCLUSION

Regardless if packed or not, trimming of roots does not affect visual quality and shelf life of UWS at 29±1.0ºC and 20±0.5ºC except for the unpacked UWS. Trimming the roots and storing UWS unpacked at 20±0.5ºC extends the shelf life by 1 day.

Wilting was significantly delayed in samples packed in PEB with 1 pinprick compared to the unpacked ones in all storage temperatures. Packing in PEB with 1 pinprick extends the shelf life by 1 day at 29±1.0ºC (with or without roots), 2 days at 20±0.5ºC (without roots), 1 day at 20±0.5ºC (with roots) and 4 days 10±0.5ºC (without roots).

Yellowing was delayed for 1, 2 and 3 days in 29±1.0ºC, 20±0.5ºC and 10±0.5ºC, respectively. This shows the effectiveness of low temperature storage in delaying the incidence of yellowing. On the other hand, no significant trends were observed on the response of UWS to MAP and presence of roots in relation to yellowing.

Disease incidence was not observed on all samples. This can be attributed to the systematic preparation of samples including washing with 100ppm hypochlorite solution and proper air drying.

Given that the preparation of leaves in this experiment was followed, packing UWS without roots in PEB with 1 pinprick in combination with storage at 10±0.5ºC extends the shelf life to 5 days, with notable delay in occurrence and reduction of the extent of wilting and yellowing.

ACKNOWLEDGEMENT

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Oral Session | Postharvest/Food Technology and Process Engineering

[5-1015-C] Postharvest/Food Technology and Process Engineering (5)

Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[5-1015-C-05] Investigation of Cowpea Variety and Storage Methods on Cowpea Beetle Infestation

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Keywords: cowpea beetle , polyethylene , Hessian bags , phostoxin , Aluminum bins

Investigation of effect of variety and storage methods on cowpea beetle (callosobruchus maculatus) infestation was carried out with the main aim of providing suitable, safe and affordable methods of storing various varieties of cowpea devoid of infestation. Three varieties of cowpea which are White, Brown and Black varieties were used. Various storage methods which include Plastics, Polyethylene, Hessian bags and Aluminum Bins of 10 kg capacity each were used in storing the cowpea. Twenty (20) cowpea beetle were introduced into the stored cowpea. Storage chemicals (Protoxin and Atelic dust) were put into the various stored cowpea at the same time of introducing the chemicals. The period of storage was four months. Completely randomized design (CRD) with four treatment and three replications were used for the experiment. Data collected include number of dead beetles, number of live beetle and number and percentage of damaged seed. The data were analyzed using analysis of variance (ANOVA). The result showed that Phostoxin and Atelic dust are toxic to the beetle in all the storage methods used leading to high mortality of the beetle though with less significant difference in the Hessian bag storage method. The result also showed that there is a strong significant difference among the treatment on White and Black varieties and no significant difference among the treatment on the Brown variety in terms of cowpea beetle damage. It was also found that Plastics and Polyethylene method of storage impaired respiration of the beetle leading to high mortality than in the Aluminum bin and the Hessian bag storage methods. Plastics and Polyethylene are therefore recommended for cheaper and environmentally safer for storage of cowpea. Also, the percentage of damage in the Black and White varieties were negligible and the period of storage with less infestation was longer than in the Brown variety.

Key words: cowpea beetle, polyethylene, Hessian bags, phostoxin, Aluminum bins.

1.0   Introduction

Cowpea (*vignaunguiculata*) (L) walp is a warm weather crop that is well adapted to drier region of the tropics like Nigeria where other food legumes do not thrive well. It is one of the most economically and nutritionally important indigenous African grain legumes produced throughout the tropical and subtropical areas of the world (Golob et. al., 1999). Nigeria is its largest producer and consumer, accounting for about 45 percent of its world production (Degri, 2008), while Africa account for about 75% (Brternburg et.al., 1995). Cowpea seed pods are consumed in fresh form as green vegetables in some African countries, while the rest of the cowpea plant serves as a nutritious fodder for livestock and also as a source of cash income when sold to farmers who use them as livestock feed. Cowpea seeds are also a rich source of minerals and obtains (Adeduntan et.al., 1998). Cowpea is sometimes called poor man meat or vegetable meat due to its high protein content. Cowpea grain contain 23.4% protein, 1.8 % fat and 60.3 % carbohydrates and also a good source of vitamins and phosphorus (Adediran and Akinneye. 2004).

In spite of the great value of cowpea particularly in Nigeria, their availability and utilization have been impaired due to the seed damage by insect pest particularly the larvae of cowpea beetle (*callosobuchusmaculatus)* (Ofuya and Lale, 2001). Attack by insect pest species begins in the field and continues in storage causing substantial damage to store grain legumes as the pest rapidly increase. It has been reported that both quantitative and qualitative losses arising from physical, chemical and biological factors e. g fungi, rodents birds and insect occur during storage of grains (Emeasor *et.al*., 2007). *Callusobruchusmaculatus*. Up to 100% infestation of cowpea can occur after three to six months storage (Maina, 2011).

Majority of farmers in Northern Nigeria and some other countries, including the Sudan, (Baribusta et. al, 2010) use local or indigenous storage facilities to forestall the menace of these insect pest they use storage insecticide where available and affordable like the banned and highly restricted lindens (gammalin A) and the acceptable are like Aluminum or Atelic EC for storing their legume grains against cowpea beets, termites, rats and disease pathogens (Degri,2007).

Some local plants have been studied to show they have effect against the activity of insect pest. They include; *NeemAzadiracta* (A.juss), Nicotine (*Nicotinia*spp), pyrethrum *chrysantheumceneraefolium*), Rotenme (*Derriselliptica*) (C.P.F, 1987). Sadim apple "Locally name Usher" (Calotropis*procera* (J.), Sesame (*Sesamumindicum* L.), Garlic (*AllumSativum* L.) and (Lantana Camara), (Mueller et. al., 1995). They were all found to lower fecundity per female and adult emergence (Singh et'al, 1996). But the availability and side effects of these are also a major concern to farmers. Hermetic storage technology has emerge as a potent alternative to other method of storage that protects commodities from insect and moulds have been developed and applied and they abound in type and the PICS (Purdue Improved Cowpea Storage) which was founded by the Bill and Melinda Gates foundation, is just one of these. The goal of the

project is to have 50 % of farm-stored cowpea in hermetic storage without insecticide in west and central Africa (Murdock et al., 2003). This is still on-going.

From the forgoing, some methods of cowpea beetle control abound but not without so many limitations, they are not cheap and some are also hazardous to health. Application of storage chemicals are sometimes not done properly by the local farmer which can lead to food poisoning. Larger quantity of cowpea are sold off immediately after harvest by the local farmers because of lack of adequate storage methods and fear of infestation by cowpea beetles thereby selling at a lower price compared to cost of production. This makes the produce scarce after the period of harvest.

This research was carried out to investigate the effect of variety and storage methods on the control of the cowpea beetle *Callosobruchus maculates (f) (coleopteran: Bruchide)* on stored cowpea. Effect of various storage methods on the control of cowpea beetle was also investigated as well as the variety that responds well to the various storage methods.

2.0   Materials and Methods

2.1   Sample collection and preparation

The following materials were used for the research, three varieties of cowpea: white variety (Kanannado), brown (Ife brown) and black (Akidi) variety. Insect pest cowpea beetle *callosobruchus maculates*, was used as the insect pest, which were introduced to each treatment at same level. The seed scanner also known as dianophoscope was used to scan the cowpea seed in order to detect the effect of insect damage from each treatment. The storage methods used in this research are polyethylene (hermetic), storage bins which are made of aluminum, plastic containers and hessian bags. The storage chemicals that were used are phostoxin and atelic dust. These chemicals were chosen because they are mostly used by farmers in Bauchi State and in the wrong proportion and application. All the experimental materials were purchased from a local grain market in Bauchi State, Nigeria.

2.2   Methods

2.2.1   Cleaning and Determination of Moisture content

The purchased cowpea were cleaned to remove debris and all other foreign materials, this was done by hand picking, sorting and using winnower. Moisture content of each of the cowpea variety were determined using standard methods as used by Abodenyi et. al., 2018. This was to ensure that the sample were at the safe storage moisture content to minimize spoilage during storage period.

2.2.2 Experimental procedures

2 kg of each variety were put in nine Polyethylene bags, the first three had phostoxine tablets introduced into it, and the next three had the atelic dust of 2 gm introduced into them, the last three served as control with no treatment. Each of the storage samples had Twenty (20) cowpea beetles introduced into them. These methods were repeated for the Aluminum storage bins, the Plastic containers and the Hessian bags for each variety. After introduction of the storage pest, the samples were agitated for one minute each to allow even spread of the pest and storage chemical (Ebiamadon et al., 2011)

The experimental set up were laid out in a completely randomized design with three replicates kept in the post-harvest laboratory of the department of agricultural bio-environmental engineering of federal polytechnic, Bauchi, Nigeria at 31 ± 2 °C and a relative humidity of 65 ± 5 for a period of 90 days

2.3 Data Collection and Statistical Analysis

The rate of infestation was determined for each variety after 90 days of infestation with the pest, the following data were collected.
1. Number of live and dead insects: this was counted manually and recorded from each treatment.
2. Percentage damage grains. The number of grains with holes and grains without roles in all the treatments in each variety: this was done by pouring the seed on a seed scanner to detect the damage seeds in each treatment, and manually counting the number of grains with holes and those without holes. The holes on the grain was used as an indicator of damage. Percentage grain damage was determined using the following formula.

$$percentage\ damage\ (\%) = \frac{number\ of\ damage\ grains}{total\ number\ of\ grain\ sampled}\ X100$$

Minitab statistical software was used in the analysis of variance (ANOVA) to determine the variation in results of all the experiments under the various independent variables and their interaction at 95% level. Descriptive statistics such as percentage was also used in presenting the data.

3.0   RESULTS

The results obtained are as presented in the tables below for the three varieties of cowpea

Table 1: Mean Effect of Cowpea Beetle Mortality on White Variety at 90 Days after Infestation

Treatments									
Storage methods	**Phostoxine**			**Atelic**			**Control**		
	Number of live beetles	Number of dead beetles	Percentage mortality (%)	Number of live beetles	Number of dead beetles	Percentage mortality (%)	Number of live beetles	Number of dead beetles	Percentage mortality (%)
Polyethylene Bags	1	19	95	3	17	85	7	13	65
Aluminum Bins	4	16	80	5	15	75	10	10	50
Hessian Bags	7	13	65	10	10	50	20	0	0
Plastic containers	0	20	100	3	17	85	6	14	70

Table 2: Mean Effect of Cowpea Beetle Mortality on Brown Variety at 90 Days after Infestation

Storage methods	Treatments								
	Phostoxine			Atelic			Control		
	Number of live beetles	Number of dead beetles	Percentage mortality (%)	Number of live beetles	Number of dead beetles	Percentage mortality (%)	Number of live beetles	Number of dead beetles	Percentage mortality (%)
Polyethylene Bags	3	17	85	5	15	75	9	11	55
Aluminum Bins	6	14	70	6	14	70	15	5	25
Hessian Bags	9	11	55	11	9	45	20	0	0
Plastic containers	3	17	85	4	16	80	9	11	55

Table 3: Mean Effect of Cowpea Beetle Mortality on Black Variety at 90 Days after Infestation

Storage methods	Treatments								
	Phostoxine			Atelic			Control		
	Number of live beetles	Number of dead beetles	Percentage mortality (%)	Number of live beetles	Number of dead beetles	Percentage mortality (%)	Number of live beetles	Number of dead beetles	Percentage mortality (%)
Polyethylene Bags	0	20	100	1	19	95	7	13	65
Aluminum Bins	3	17	85	5	15	75	11	9	45
Hessian Bags	7	13	65	8	12	60	20	0	0
Plastic containers	0	20	100	2	18	90	7	13	65

Table 4: Mean Percentage (%) of Damaged Cowpea at 90 Days after Infestation

	Treatments								
Storage methods	**White variety**			**Brown variety**			**Black variety**		
	Phostoxine	Atelic	Control	Phostoxine	Atelic	Control	Phostoxine	Atelic	Control
Polyethylene Bags	10	12	40	15	15	50	8	10	30
Aluminum Bins	40	50	60	50	55	60	30	40	55
Hessian Bags	50	60	90	60	60	90	40	50	70
Plastic containers	9	10	35	12	20	40	8	10	30

3.1   Discussion

3.1.1   Cowpea Beetle Mortality on the various varieties of cowpea

The control treatment was generally less effective than the phostoxine and atelic dust at 90 days of storage and infestation of the cowpea. From tables 1, 2 and 3 all the storage methods were effective against the insect with significantly varying degree of efficiencies. Cowpea beetle mortality was significantly affected on the white variety more especially on the polyethylene storage and plastic containers with 95% and 100% mortality respectively. The Atelic showed mortality rate of 85 % for both polyethylene and plastic containers while the control treatment has a value of 65 % and 70 % for both the polyethylene and plastic containers respectively. The Aluminum bin showed 80% mortality on phostoxine combination, 75% for the atelic and 50 % for the control treatment. The reduction of oxygen during the 90 days of storage after the infestation reduced the insect count drastically especially in the polyethylene bags and the plastic containers. This cannot be said of the Hessian bags because they are porous and allowed the

thriving of the storage pest in all the treatments. This result agrees with the findings of (Ebiamadon et al., 2011) which researched the effectiveness of different botanical pesticides on control of *C. maculatus* at 30 and 90 days of infestation.

The mortality of cowpea beetle on the Brown variety, cowpea beetle mortality was significantly affected by the storage chemicals and the storage methods. Polyethylene together with phostoxine and plastic containers showed high mortality of 85 %. Atelic with polyethylene and plastic containers has mortality rate of 75 % and 80 % respectively. The control treatment indicated mortality of 55 %, this result agrees with PICS project (Villers, et al., 2008) which used the Hermetic storage methods by keeping away oxygen from the pest they were able to record 50 % mortality. The Hessian bags showed 0 % mortality for the control treatment.

Cowpea beetle mortality on the Black variety was significantly high after the 90 days infestation and storage for all the treatments and storage methods except for the Hessian bags that indicated 65 % for phostoxine, 60 % for atelic and 0 % for the control treatment.

3.1.2 Cowpea Damage at 90 Days after Infestation

Table four shows the degree of damage on the three varieties of cowpea after 90 days of infestation. The Hessian bag recorded the highest percentage of damage on all the storage methods and treatments for the three varieties. This can be attributed to the fact that the Hessian bag is porous that allowed intake of oxygen that allowed the survival of the storage pest. The White and black variety recorded less damage form the beetle from all the storage methods and treatments, this could be as a result of the high protein content of Brown beans, storage pest tend to fist more on highly protein food (AOAC, 2010)

4.0 Conclusion

From the above results, it can be concluded that the black variety is less susceptible to cowpea infestation when stored in a polyethylene bag as well as in an airtight plastic container.

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Oral Session | Others (including the category of JSAM and SASJ)

[5-1015-D] Other Categories (2)

Chair:Tri Yuliana(Universitas Padjadjaran, Indonesia)
Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room D (4th room)

[5-1015-D-01] **Screening and Enzyme Activity of Cellulose-Producing Bacteria Isolated from Kemiri Sunan (*Reutealis trisperma* (Blanco) Airy Shaw) and Empty Fruit Bunches of Palm Oil**

*Tri Yuliana[1], Efri Mardawati[1], Souvia Rahimah[1], Emilda Ayu Febrianty[1], Agus Try Hartono[1] （1. Univ. Padjadjaran, Indonesia(Indonesia)）

10:15 AM - 10:30 AM

[5-1015-D-02] **Development of a Cloud-based Internet of things Monitoring System for Fish Activity and Water Quality in Aquaponics**

*Chien Lee[1], Yu-Jen Wang[1] （1. Department of Mechanical and Electromechanical Engineering, National Sun Yat-sen University(Taiwan)）

10:30 AM - 10:45 AM

[5-1015-D-03] **EFFECT OF DIFFERENT MODES OF PLANTING AND WEEDING ON MACHINE FIELD CAPACITY AND YIELD OF A MIXED CROPPING SMALL HOLDER FARM**

Folasayo Titilola Fayose[1], Adesoji Mathew Olaniyan[1], *Babatope Albert Alabadan[1], Anthony Ayodele Fajinmi[1], Kayode Ogunleye[1], Olanrewaju Omoju[1], Olufemi Aladejebi[1], Oluwaseun Ilesanmi[1] （1. Federal University Oye Ekiti(Nigeria)）

10:45 AM - 11:00 AM

[5-1015-D-04] **Development of Agro-industrial Worker Trust Assessment System for Sustainable Ergonomic Program in Food Small and Medium-sized Enterprises**

*Mirwan Ushada[1], Nur Achmad Sulistyo Putro[2], Titis Wijayanto[3], Fitri Trapsilawati[3], Nafis Khuriyati[1] （1. Universitas Gadjah Mada, Department of Agro-industrial Technology(Indonesia), 2. Universitas Gadjah Mada, Department of Computer Science and Electronics(Indonesia), 3. Universitas Gadjah Mada, Department of Mechanical and Industrial Engineering(Indonesia)）

11:00 AM - 11:15 AM

[5-1015-D-05] **ASSESSING LAND USE TYPES IMPACT ON SOIL ORGANIC CARBON IN SOUTH WEST, NIGERIA**

*OLORUNWA ERIC OMOFUNMI[1], ADESOJI MATTHEW OLANIYAN[1] （1. FEDERAL UNIVERSITY OYE-EKITI(Nigeria)）

11:15 AM - 11:30 AM

Oral Session | Others (including the category of JSAM and SASJ)
[5-1015-D] Other Categories (2)
Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room D (4th room)

[5-1015-D-01] **Screening and Enzyme Activity of Cellulose-Producing Bacteria Isolated from Kemiri Sunan (*Reutealis trisperma* (Blanco) Airy Shaw) and Empty Fruit Bunches of Palm Oil**

*Tri Yuliana[1], Efri Mardawati[1], Souvia Rahimah[1], Emilda Ayu Febrianty[1], Agus Try Hartono[1] (1. Univ. Padjadjaran, Indonesia(Indonesia))
Keywords: cellulose, Reutealis trisperm, Palm bunches, clear zone, enzyme activity

Biocatalyst technology is needed for the industry to improve performance of production. Cellulase enzymes has an important role in biocatalyst technology, especially in pulp industry. Cellulase is produced by certain types of microbes. The selection of cellulase-producing bacteria from Trisperma shell and empty fruit bunches from oil palm were carried out in order to produce cellulase which can be used for the pulp industry. Effectiveness test of cellulase-producing bacteria from Trisperma shell and palm bunches were also carried out using the liquid phase fermentation method. The result shows isolat K2 gave the widest clear zone with a value of 77.19% ± 0.00835 in BSM-CMC-CR media. OD value was calculated within 8 hours, 24 hours, 32 hours, and 48 hours in NB media. The result shows at 32 hours, the K3 isolate gave the highest absorbance with the value of 0.9163. Test of enzyme activity shown the K3 isolate had a highest enzyme activity with its value of 43.2 x 10-5 U/mL at 48 hours. The result of gram negative bacteria staining was assumed that the bacteria was *Pseudomonas* sp.

Screening and Enzyme Activity of Cellulose-Producing Bacteria Isolated from Kemiri Sunan (*Reutealis trisperma* (Blanco) Airy Shaw) and Empty Fruit Bunches of Palm Oil

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ABSTRACT

Biocatalyst technology is needed for the industry to improve performance of production. Cellulase enzymes has an important role in biocatalyst technology, especially in pulp industry. Cellulase is produced by certain types of microbes. The selection of cellulase-producing bacteria from Trisperma shell and empty fruit bunches from oil palm were carried out in order to produce cellulase which can be used for the pulp industry. Effectiveness test of cellulase-producing bacteria from Trisperma shell and palm bunches were also carried out using the liquid phase fermentation method. The result shows isolat K2 gave the widest clear zone with a value of $77.19\% \pm 0.00835$ in BSM-CMC-CR media. OD value was calculated within 8 hours, 24 hours, 32 hours, and 48 hours in NB media. The result shows at 32 hours, the K3 isolate gave the highest absorbance with the value of 0.9163. Test of enzyme activity shown the K3 isolate had a highest enzyme activity with its value of 43.2×10^{-5} U/mL at 48 hours. The result of gram negative bacteria staining was assumed that the bacteria was *Pseudomonas* sp.

Keywords: *Reutealis trisperm, Palm bunches, cellulose, clear zone, enzyme activity*

10:30 AM - 10:45 AM (Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room D)

[5-1015-D-02] Development of a Cloud-based Internet of things Monitoring System for Fish Activity and Water Quality in Aquaponics

*Chien Lee[1], Yu-Jen Wang[1] （1. Department of Mechanical and Electromechanical Engineering, National Sun Yat-sen University(Taiwan)）
Keywords: Aquaponics, Aquaculture, Internet of Things, Fish Activity, Oxygen Transfer, Water Quality

A cloud-based Internet of things monitoring system in aquaponics is proposed in this study. The system can use commercial sensors to measure water temperature, water depth, the amount of oxygen dissolved in water, and water pH. Moreover, three infrared distance sensors were attached to the aquarium glass at different heights to estimate fish school activity and provide an alternative alarm system for indicating an abnormal water level in the tank. Water depth sensing in the rearing tank can be used to evaluate the ebb-and-flow irrigation function and estimate the flow rate of water circulation. A novel oxygen transfer model was set up in this study, the results of which prove that fish activity influences water quality. The model also indicates how to use regression analysis for diagnosing problems. Fish activity measurements can be used to estimate water quality or cross-check sensor types and provide proactive precursors to variations. The measuring module containing sensors and sub-1 GHz communication can transmit data through a 1-km-long gateway module. Finally, the data are uploaded to ThingSpeak[TM], a cloud platform, through Wi-Fi. By using the data stored on the cloud, a real-time alarm system for indicating abnormalities is developed and a periodic regression analysis is conducted using the cloud-based programming of ThingSpeak[TM].

10:45 AM - 11:00 AM (Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room D)

[5-1015-D-03] EFFECT OF DIFFERENT MODES OF PLANTING AND WEEDING ON MACHINE FIELD CAPACITY AND YIELD OF A MIXED CROPPING SMALL HOLDER FARM

Folasayo Titilola Fayose[1], Adesoji Mathew Olaniyan[1], *Babatope Albert Alabadan[1], Anthony Ayodele Fajinmi[1], Kayode Ogunleye[1], Olanrewaju Omoju[1], Olufemi Aladejebi[1], Oluwaseun Ilesanmi[1] （1. Federal University Oye Ekiti(Nigeria)）
Keywords: planting, weeding, field capacity, yield

Nigeria has great potential for cultivation of a wide variety of crops as its soil and climatic conditions are suitable for crop cultivation. However, growing crops with human labour (planting, weeding) has been the common practice. After an initial conventional tillage, labour saving mechanical jab and rotary planters, reciprocating weeder and manual methods were used to establish a mixed cropping one hectare farm of maize and cassava under rain-fed conditions The effects of these treatments were studied using the following parameters: field capacity of planting, weeding and yield of crops. The highest field capacity among the planting modes was that of rotary planting with 1.53 ha/hr while, 0.44 ha/hr and 0.24 ha/hr were obtained for jab and manual planting respectively. A field capacity of 0.012ha/hr was obtained for mechanical weeding as against 0.0036 ha/hr with manual weeding. The yields of the maize stover are as follows: Manual planting 6.9 tonnes/ha, Rotary planting 11.5 tonnes/ha, Jab planting 3.9 tonnes/ha while that of the average ear weight are 15.42 tonnes/ha for rotary planting, 10.33 tonnes/ha for manual planting and 5.83 tonnes/ha for jab planting. The effect of the use of chemical weeding reduced the yield of cassava roots to 60 ton/ha as

against 81 ton/ha for manual/mechanical weeding. Further investigation is ongoing to substantiate the facts. However, these observations are in agreement with the fact that mechanical manipulation of the soil by way of planting and weeding loosen the soil between rows, thus increasing air and water intake capacity, thereby increasing yield.

11:00 AM - 11:15 AM (Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room D)

[5-1015-D-04] Development of Agro-industrial Worker Trust Assessment System for Sustainable Ergonomic Program in Food Small and Medium-sized Enterprises

*Mirwan Ushada[1], Nur Achmad Sulistyo Putro[2], Titis Wijayanto[3], Fitri Trapsilawati[3], Nafis Khuriyati[1]　(1. Universitas Gadjah Mada, Department of Agro-industrial Technology(Indonesia), 2. Universitas Gadjah Mada, Department of Computer Science and Electronics(Indonesia), 3. Universitas Gadjah Mada, Department of Mechanical and Industrial Engineering(Indonesia)）
Keywords: Bird swarm algorithm, Collective trust, Environmental ergonomics, Individual trust, Kansei Engineering

Ergonomic program has not yet fully gained the worker trust in food Small Medium-sized Enterprises (SMEs) due to the gap between ergonomics and financial amenities. The tangible financial amenities as wages, incentives, and insurance have been more attractive than the intangible ergonomics program in the form of a comfortable workplace environment (Environmental ergonomics), efficient work methods and optimum workload. Trust could be defined as an abstractive (Kansei) human factor which is characterized by uncertainty and vulnerability to support their individual and collective decision. Trust influence the attractiveness of ergonomic program to worker as individual and worker union as the collective. The abstractive communication between 1 (one) individual worker and other partners in same union is possible to be simulated in an artificial bird swarm algorithm. Kansei engineering was selected to model the individual trust due to the reliability for modeling the abstractive human factors. Artificial swarm intelligence was selected to simulate the collective trust due to capability to model non-linear of human factors. The research goal was to develop an agro-industrial worker trust assessment system for sustainable ergonomic program in food SMEs. The research objective was: 1) To predict the worker individual trust using Kansei Engineering; 2) To simulate the worker collective trust using bird swarm algorithm. The system is expected to assist the SME's management for developing trust evidence-based ergonomic policy. Generally, the system is expected to support the Sustainable Development Goals numbers 3 (Good health &well-being) and number 9 (Industry, innovation and infrastructure). The system was tested on the database of worker human factors in Food SMEs. The inputs of the system was extracted from database as: 1) Workload; 2) Workplace temperature; 3) Relative humidity; 4) Light intensity; 5) Incentive. The output was individual and collective trust. The agro-industrial worker trust assessment system consists of 7 sub-systems. In the Sub-system 1, measurement is carried out to obtain the worker mood states, heart rate and workplace environment parameters. In Sub-system 2, the manager obtain measurement result in Sub-system 1 as the input to determine integrated workload and workplace temperature set point. If the workload indicated the normal status, then the workplace temperature is set. If the workload status indicated under or over load, then the system provides feedback for the manager to evaluate the existing ergonomic program. In Sub-system 3, the temperature was set in an air conditioner to create the comfortable workplace environment (Environmental ergonomics). In Sub-system 4, the work incentive is determined based on integrated workload (Sub-system 2) and environmental ergonomics (Sub-system 3). The individual trust index is determined in Sub-system 5. If

the index indicated the status of trust, the system proceeds the status to the Sub-system 6. If the index indicated distrust, the system provides feedback to the manager to evaluate the existing ergonomic program. The Sub-system 6 processes the individual trust in Sub-system 5 using the Bird Swarm Algorithm in Kansei Engineering (BISAKE). The algorithm simulated the worker union to behave like a bird swarm in determining whether an individual trust is satisfied or not against their mentality constraints of prior knowledge, familiarity, agreement and preference. Finally, in the Sub-system 7, the collective trust was validated. The simulation result indicated that worker trust index could be assessed based on workload status, a percentage of incentive and workplace environmental cost per month. Furthermore, this assessment could make the trust data more manageable to store, retrieve and enable interchange in big data system for sustainable ergonomic program in food SMEs.

Oral Session | Others (including the category of JSAM and SASJ)
[5-1015-D] Other Categories (2)
Thu. Sep 5, 2019 10:15 AM - 11:30 AM Room D (4th room)

[5-1015-D-05] ASSESSING LAND USE TYPES IMPACT ON SOIL ORGANIC CARBON IN SOUTH WEST, NIGERIA

*OLORUNWA ERIC OMOFUNMI[1], ADESOJI MATTHEW OLANIYAN[1] （1. FEDERAL UNIVERSITY OYE-EKITI(Nigeria)）
Keywords: Federal University Oye Ekiti (Ikole campus), land use type, Soil organic carbon, Soil properties

The amount of soil organic carbon (SOC) stored in a particular soil is influenced by several factors including climate, vegetation type, land management, soil properties and current and last land use. The impacts of land use types on soil organic carbon were assessed. Four land use types were used in the study. Sampled soils were tahen at depth of 0 - 45 cm and at intervals of 15 cm. The soil samples were examined in accordance with the standard methods described by the American Public Health Association (APHA). The data were analyzed using descriptive statistics. The results showed the mean soil organic carbon content was higher under oil palm plantation land [D] compared with ther land use types at 0 - 15 cm soil depth (22.87g/kg) which was 1.5, 2.6 and 53.3 % more than in the Faculty of Agriculture Teaching and Research farm land [A], the cashew plantation land [B] and the Agricultural and Bioresources experimental farm land [C] respectively. This could be attributed to the greater inputs of vegetation (litter fall) and reduced decomposition of organic matter. Similarly, the lowest soil organic carboncontent under land use type C could be due to reduced inputs of organic matter and frequent tillage which encouraged oxidation oforganic matter. The finding indicated that the means of SOC in land use types were no significantly different (p = 0.05) except in the land use type C. It is concluded that land use types have influenced on soil organic carbon

ASSESSING LAND USE TYPES IMPACT ON SOIL ORGANIC CARBON IN SOUTH WEST, NIGERIA

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ABSTRACT

The amount of soil organic carbon (SOC) stored in a particular soil is influenced by several factors including climate, vegetation type, land management, soil properties and current and past land use. The impacts of land use types on soil organic carbon were assessed. Four land use types were used in the study. Sampled soils were taken at depth of 0 – 45 cm and at intervals of 15 cm. The soil samples were examined in accordance with the standard methods described by the American Public Health Association (APHA). The data were analyzed using descriptive statistics. The results showed that mean soil organic carbon content was higher under the oil palm plantation land use [D] compared with other land use types at 0 – 15 cm soil depth (22.87 ± 3.89 g kg^{-1}), which was 1.5, 2.6 and 53.3 % more than in the Faculty of Agriculture Teaching and Research farm land A], the cashew plantation land [B] and the Agricultural and Bioresources experimental farm land [C] respectively. This could be attributed to greater inputs of vegetation (litter fall) and reduced decomposition of organic matter. Similarly, the lowest soil organic carbon content under land use type C could be due to reduced inputs of organic matter and frequent tillage which encouraged oxidation of organic matter. The finding indicated that the means of SOC in land use types were no significantly different (P = 0.05) except in the land use type C. It is concluded that land use types have influenced on soil organic carbon

Keywords: Federal University Oye Ekiti (Ikole campus), land use type, Soil organic carbon, Soil properties

Oral Session | Food Function/Nutrition

[6-1015-A] Functional/Wellness Foods &Nutrition (2)

Chair:Rungarun Sasanatayart(Mae Fah Luang University, Thailand)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Hall A (Main Hall)

[6-1015-A-01] **Change of Bioactive Compounds and Bioactivities of Crisphead Lettuce during Simulated *In Vitro* Digestion**
*Sunantha Ketnawa[1], Yukiharu Ogawa[1] （1. Graduate School of Horticulture, Chiba University(Japan)）
10:15 AM - 10:30 AM

[6-1015-A-02] **Impact of Crystallinity Change During *In Vitro* Digestion on Starch Digestibility of Microwave- and Steam-Cooked Black Rice**
*Sukanya Thuengtung[1], Yoshitaka Matsushita[2], Yukiharu Ogawa[1] （1. Graduate School of Horticulture, Chiba University(Japan), 2. Research Network and Facility Services Division, National Institute for Materials Science (NIMS)(Japan)）
10:30 AM - 10:45 AM

[6-1015-A-03] **Study of Static *In Vitro* Digestion of Japanese Pickled Plums on the Change of Polyphenols and Antioxidant Activity**
*Jutalak Suwannachot[1], Sunantha Ketnawa[1], Yukiharu Ogawa[1] （1. Chiba University(Japan)）
10:45 AM - 11:00 AM

[6-1015-A-04] **Preparation of Pigment Extract Loaded Alginate Beads and Their Stability of Antioxidant Activities during *in vitro* Gastrointestinal Digestion**
*Rungarun Sasanatayart[1], Sutthiwal Setha[1] （1. School of Agro-Industry, Mae Fah Luang University(Thailand)）
11:00 AM - 11:15 AM

[6-1015-A-05] **Stability of Plant Pigments and Antioxidant Activities in Juice Model during Processing and *in vitro* Gastrointestinal Digestion**
*Titikan Liangpanth[1], Rungarun Sasanatayart[1] （1. School of Agro-Industry, Mae Fah Luang University(Thailand)）
11:15 AM - 11:30 AM

Oral Session | Food Function/Nutrition
[6-1015-A] Functional/Wellness Foods &Nutrition (2)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Hall A (Main Hall)

[6-1015-A-01] Change of Bioactive Compounds and Bioactivities of Crisphead Lettuce during Simulated *In Vitro* Digestion

*Sunantha Ketnawa[1], Yukiharu Ogawa[1] (1. Graduate School of Horticulture, Chiba University(Japan))
Keywords: In vitro digestion, crisphead lettuce, bioactive compounds, bioactivity, bioaccessibility

Lettuces (*Lactuca sativa* L.) are the most popular vegetables in the world and are consumed in increasing amounts due to a good contribution to human health. Favorite species are butterhead, romaine, and, most importantly, crisphead (iceberg) lettuce. Crisphead lettuce is also of particular interest due to its high content in antioxidants and phytochemicals including caffeic acid and its derivatives, flavonols, vitamins C and E, chlorophyll, and carotenoids. The potential health benefit of crisphead lettuce before and after the simulated *in vitro* digestion will be represented by the recovery, bioaccessibility, and change of bioactive compounds [including total phenolic (TPC) and total flavonoids content (TFC)] and bioactivities [*in vitro* antioxidant activities including 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2, 2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radical scavenging activities, ferric reducing antioxidant power (FRAP) and metal ion chelating (MIC)]. Thus, the objectives of this study were to study the change on recovery and bioaccessibility of those properties of cripshead lettuce utilizing simulated *in vitro* digestion model. The results suggest that the releasing of bioactive compounds as well as bioactivities increased during gastric digestion and intestinal digestion for 1 h then decreased when IVD completed, thus coordinated with recovery and bioaccessibility index. Crisphead lettuce showed the highest recovery and bioaccessibility of TPC and TFC at gastric phase digestion for more than 60% and 70%, respectively whereas the lowest of those found in after finish digestion for around 50% for both TPC and TFC. Among all bioactivities, crisphead lettuce sample showed the recovery and bioaccesibility of ABTS (61-92%) followed by FRAP (71-84%), DPPH (24-52%) and MIC (21-46%) during the digestion. The present study suggests that cripshead lettuce maintains stability in both bioactive compound and bioactivities during the digestion. Regarding the 4 methods used, significant correlations were found between bioactive compounds and antioxidant activity while ABTS exhibited weaker with TFC than TPC. Taken together, a relatively tight coupling of four parameters indicates that every one of them can be considered as a relevant and reliable characteristic of the antioxidant capacity of lettuce. This would provide a scientific basis for further studies on develop formulating new functional foods due to high nutritional value tolerant.

Change of Bioactive Compounds and Bioactivities of Crisphead Lettuce during Simulated *In Vitro* Digestion

Sunantha Ketnawa and Yukiharu Ogawa

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ABSTRACT

Lettuces (*Lactuca sativa* L.) are the most popular vegetables in the world and are consumed in increasing amounts due to a good contribution to human health. Favorite species are butterhead, romaine, and, most importantly, crisphead (iceberg) lettuce. Crisphead lettuce is also of particular interest due to its high content in antioxidants and phytochemicals including caffeic acid and its derivatives, flavonols, vitamins C and E, chlorophyll, and carotenoids. The potential health benefit of crisphead lettuce before and after the simulated *in vitro* digestion will be represented by the recovery, bioaccessibility, and change of bioactive compounds [including total phenolic (TPC) and total flavonoids content (TFC)] and bioactivities [*in vitro* antioxidant activities including 2,2-diphenyl-1-picrylhydrazyl (DPPH), 2, 2'-azinobis-(3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) radical scavenging activities, ferric reducing antioxidant power (FRAP) and metal ion chelating (MIC)]. Thus, the objectives of this study were to study the change on recovery and bioaccessibility of those properties of cripshead lettuce utilizing simulated *in vitro* digestion model. The results suggest that the releasing of bioactive compounds as well as bioactivities increased during gastric digestion and intestinal digestion for 1 h then decreased when IVD completed, thus coordinated with recovery and bioaccessibility index. Crisphead lettuce showed the highest recovery and bioaccessibility of TPC and TFC at gastric phase digestion for more than 60% and 70%, respectively whereas the lowest of those found in after finish digestion for around 50% for both TPC and TFC. Among all bioactivities, crisphead lettuce sample showed the recovery and bioaccesibility of ABTS (61-92%) followed by FRAP (71-84%), DPPH (24-52%) and MIC (21-46%) during the digestion. The present study suggests that cripshead lettuce maintains stability in both bioactive compound and bioactivities during the digestion. Regarding the 4 methods used, significant correlations were found between bioactive compounds and antioxidant activity while ABTS exhibited weaker with TFC than TPC. Taken together, a relatively tight coupling of four parameters indicates that every one of them can be considered as a relevant and reliable characteristic of the antioxidant capacity of lettuce. This would provide a scientific basis for further studies on develop formulating new functional foods due to high nutritional value tolerant.

Keywords: *In vitro* digestion, crisphead lettuce, bioactive compounds, bioactivity. bioaccessibility

10:30 AM - 10:45 AM (Fri. Sep 6, 2019 10:15 AM - 11:30 AM Hall A)

[6-1015-A-02] Impact of Crystallinity Change During *In Vitro* Digestion on Starch Digestibility of Microwave- and Steam-Cooked Black Rice

*Sukanya Thuengtung[1], Yoshitaka Matsushita[2], Yukiharu Ogawa[1] (1. Graduate School of Horticulture, Chiba University(Japan), 2. Research Network and Facility Services Division, National Institute for Materials Science (NIMS)(Japan))
Keywords: Black rice, Crystallinity, Microwave-cooking, Steam-cooking, Starch digestibility, X-ray diffraction

Rice is a major cereal crop that is consumed as staple food by around half of the world's population. Rice can be distinguished by the color of grain's pericarp layer, as pigmented and non-pigmented rice. Black pigmented rice has been long cultivated in Southeast Asian countries, and has received much attention due to its nutritional values. Carbohydrate is known as a major macronutrient of rice that could be accounted as starch content for 90%. Starch digestion is a complex process that occurs mainly in the small intestine. Several factors could influence the starch digestibility such as starch granule structure, particle size, and food processing. Consequently, the effect of microwave and steam-cooking on change of crystallinity during *in vitro* digestion, including their impact on starch digestibility of black rice were examined. Influence of rice attribute on starch digestibility was also investigated. In this study, unpolished Thai black rice (cv. Hom Nin) was soaked at 10 º C for 19 h before cooked by microwave and steam methods for 12 and 25 min, respectively. No white core inside cooked rice grain indicated full gelatinization. A portion of cooked black rice grain was ground to obtain homogenized slurry sample. Intact rice grain and homogenized slurry samples were then transferred into each *in vitro* reactor to start the simulated digestion. There were two digestion states included simulated gastric and simulated small intestinal digestion, which the supernatant was collected during these digestive states to analyze hydrolysis of starch. In addition, rice grain samples of uncooked rice, and cooked rice before and during *in vitro* digestion, were collected and prepared as the rice flour for analysis of X-ray diffraction (XRD) pattern. The results revealed that XRD pattern of uncooked black rice exhibited the diffraction peak at 15, 17, 18, and 23º (2θ), indicating A-type crystalline structure. However, XRD pattern was changed after cooking which diffraction peak was found at 13 and 20º (2θ), indicating V_h-type crystalline structure. Moreover, XRD pattern and the degree of crystallinity of steam-cooked rice were outstandingly changed after 360 and 480 min of simulated small intestinal digestion, comparing to microwave-cooked rice. The equilibrium starch hydrolysis (C_∞) percentage of steam-cooked rice showed lower than that of microwave-cooked rice as well. Besides, structure-less of homogenized slurry sample influenced faster starch hydrolysis rate when compared with intact grain sample. This study implied that rice attribute has an impact on starch hydrolysis rate of black rice, which different C_∞ between two cooking methods could be involved with change of the crystallinity degree during *in vitro* digestion.

10:45 AM - 11:00 AM (Fri. Sep 6, 2019 10:15 AM - 11:30 AM Hall A)

[6-1015-A-03] Study of Static *In Vitro* Digestion of Japanese Pickled Plums on the Change of Polyphenols and Antioxidant Activity

*Jutalak Suwannachot[1], Sunantha Ketnawa[1], Yukiharu Ogawa[1] (1. Chiba University(Japan))
Keywords: Antioxidant activity, In vitro digestion, Pickled plum, Polyphenols

Japanese plum (Ume in Japanese; *Prunus mume*) is basically consumed in processed forms, e.g. a pickled and dried form (Umeboshi), liquored form (Umeshu), and concentrated form (Bainiku-ekisu). These products have been known to possess various medicinal benefits and have been frequently prescribed as a traditional folk medicine, owing to the fruit is a good source of organic acids, edible fiber, minerals, and phenolic compounds. Two types of pickled plums (PP) called kari-kari ume (hard type [HPP]) and umeboshi (soft type [SPP]) from commercial products in Japan were used and evaluated. These products are widely consumed with rice or processed to be puree and paste products. In this study, the static *in vitro* digestion was investigated with and without digestive enzymes (control [CT]) by sampling at undigested stage (G0), 1 hour after gastric digestion (G1), 1 hour after small intestinal digestion (G1I1) and 2 hours after small intestinal digestion (G1I2). The changes of polyphenols (total phenolics [TPC] and total flavonoid contents [TFC]) and antioxidant activities (DPPH- and ABTS-radical scavenging activities, ferric reducing antioxidant power [FRAP], and metal ion chelating [MIC] activity) were investigated during simulated *in vitro* gastrointestinal digestion. Increment of TPC and TFC was found during the simulated digestion in both of PP samples. In addition, higher antioxidant activities were found during simulated digestion of digested PP when compared to those of CT. Furthermore, higher ABTS and FRAP activities were observed in HPP during gastric stage than those SPP. Nevertheless, released antioxidant activities of SPP were better than those of HPP during the small intestinal stage, except MIC activity. Regarding the releasing of antioxidant activity may be related to cell structure. Thus, differences in the initial fruit maturity and production method of PP could enhance the releasing of antioxidant activities. In conclusion, these results provided information about developing new functional food products. Therefore, the study can be applied to develop a processing method to provide maximum bioactivity for the improvement of human well-being.

11:00 AM - 11:15 AM (Fri. Sep 6, 2019 10:15 AM - 11:30 AM Hall A)

[6-1015-A-04] Preparation of Pigment Extract Loaded Alginate Beads and Their Stability of Antioxidant Activities during *in vitro* Gastrointestinal Digestion

*Rungarun Sasanatayart[1], Sutthiwal Setha[1]   (1. School of Agro-Industry, Mae Fah Luang University(Thailand)）
Keywords: Anthocyanins , Carotenoids , Antioxidant , Alginate based Encapsulation , in vitro gastrointestinal digestion

Plant pigments are safe for food applications and exert antioxidant activities, providing health benefits superior to synthetic colorants. However, there are challenges related to color losses during food processing, storage, and commercialization due to a low stability of natural pigments. In addition, based on human gastrointestinal digestion, levels of bioactive compounds and their related antioxidant activities are significantly altered. One effective approach to preserve the health beneficial properties of plant pigments is to incorporate them into polymer matrices for the improvement of stability and bioavailability. In this study, 2.5% w/w pigment extracted from butterfly pea flower (BPF) and 5% w/w pigment extracted from turmeric rhizome (TR) were prepared. PBF and TR loaded alginate beads were prepared by extrusion method using 1%w/v alginate and 2%w/v calcium chloride ($CaCl_2$). To examine the stability through simulated gastrointestinal digestion, a certain amount of BPF and TR loaded alginate beads were either added into distilled water or formulated into juice model samples containing 10%w/v glucose and 0.1M citric acid. To assess effect of heat, all samples were adjusted to pH 3.0 and were subjected to pasteurization at 75°C for

15 min. Samples without heat treatment were also compared. Samples were examined for antioxidant properties in terms of total anthocyanins content (TAC) for BPF, total carotenoids content (TCC) for TR, total phenol content (TPC), total flavonoid content (TFC) and antioxidant activities based on FRAP and DPPH assays. During *in-vitro* gastrointestinal digestion, samples showed different amount and stability of TAC, TCC, TPC, TFC and antioxidant activities based on FRAP and DPPH, regarding type of pigments. Results showed that alginate-based encapsulation of pigment extract limited the release of bioactive compound during *in vitro* gastrointestinal digestion. The effect of either pasteurized heat alone or in combining with ingredients in releasing more bioactive components of juice model samples was also noted. During *in vitro* gastrointestinal digestion, the high stability of bioactive compounds and antioxidant activities from oral phase (G0) to gastric phase (G30) but subsequently lower stability along intestinal phase (I0-I120) were observed in all samples. These stability data supports the beneficial effect of alginate-based encapsulation in controlled releasing of bioactive compounds of plant pigments and their bioavailability which could be further applied in food industry.

11:15 AM - 11:30 AM (Fri. Sep 6, 2019 10:15 AM - 11:30 AM Hall A)

[6-1015-A-05] Stability of Plant Pigments and Antioxidant Activities in Juice Model during Processing and *in vitro* Gastrointestinal Digestion

*Titikan Liangpanth[1], Rungarun Sasanatayart[1] (1. School of Agro-Industry, Mae Fah Luang University(Thailand))
Keywords: Anthocyanins , Carotenoids , Betalains, Chlorophylls, in vitro gastrointestinal digestion, Antioxidant

Due to the changing perceptions of consumers to consume natural products, there is an increasing interest in the use of natural antioxidant pigments as substitutes for synthetic food colorants. Plant pigments exert antioxidant activities, providing health benefits. However, there are challenges related to color losses during food processing, storage, and commercialization due to a low stability of natural pigments compared to synthetic colorants. In this study, four pigment powders extracted from selected plant parts including butterfly pea flower rich in anthocyanins (TAC), dragon fruit peel containing betalains (TBC), turmeric rhizome containing curcuminoids (TCC) and pandan leaves rich in chlorophylls (TCPC) were compared. To examine the stability through processing, juice model samples containing 10% (w/v) glucose, 0.1M citric acid and coloring with 1.0-3.0% (w/v) color powder were prepared. The samples were adjusted to pH 3 and 7 and subjected to three different heat treatments including (1) No heat (control), pasteurization (75° C for 15 min) and sterilization (121° C for 15 min). All samples were measured for color parameters (L*, a*, b*, hue and Δ E) whilst, antioxidant properties were measured in terms of total phenol content (TPC) and antioxidant activities based on FRAP and DPPH assays. Results showed that pH and heat treatments played a critical role on stability of pigment compounds, resulting in change in visual color and color parameters which could limit food uses. Overall, pasteurization of juice model samples rich in plant pigments at both pH 3 and 7 was better than sterilization in retaining initial color and maximize level of bioactive compounds and related antioxidant properties. To assess stability and bioaccessibility of antioxidant properties during simulated *in-vitro* gastrointestinal digestion, all pasteurized juice model solutions at pH 3 were compared. During digestion, all juice model solutions showed different stability of pigments compound, TPC and antioxidant activities based on FRAP and DPPH. Overall, trends observed were the increased stability of bioactive compounds and their related antioxidant activities from oral phase (G0) to gastric phase (G30) but

subsequently decreased stability along intestinal phase (I0-I120). All pigment compounds, TAC, TBC, TCC and TCPC showed the less correlation with antioxidant activities based on FRAP and DPPH than did TPC. Results suggested that the concentration of pigment compounds and the antioxidant capacity before digestion might not reflect the after digested concentration. Data of this study provided relevant information on antioxidants reflecting stability and activity during digestion which supports the potential use of plant pigments in natural foods and beverages.

Oral Session | Postharvest/Food Technology and Process Engineering

[6-1015-C] Postharvest/Food Technology and Process Engineering (6)

Chair:Xujun Ye(Hirosaki University, Japan)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[6-1015-C-01] Spatially Resolved Interactance Spectroscopy to Estimate Degree of Red Coloration in Red-fleshed Apple Cultivar 'Kurenai-no-Yume'

*Xujun Ye[1], Sou Takada[1], Shuhuai Zhang[1]　(1. Hirosaki University(Japan)）

10:15 AM - 10:30 AM

[6-1015-C-02] Use of hyperspectral imaging to separate cultivars and evaluate the internal quality of nectarines

Sandra Munera[1], Prieto Andres[1], Nuria Aleixos[2], Sergio Cubero[1], *Jose Blasco[1]　(1. Centro de Agroingeniería. Instituto Valenciano de Investigaciones Agrarias (IVIA). Ctra. Moncada-Náquera Km 4.5, 46113, Moncada, Valencia(Spain), 2. Departamento de Ingeniería Gráfica. Universitat Politècnica de València. Camino de Vera, s/n, 46022 Valencia(Spain)）

10:30 AM - 10:45 AM

[6-1015-C-03] Evaluating the Performance of Unmanned Crop Sensing Robot for Rice

*Dhirendranath Singh[1], Shigeru Ichiura[1], Mitsuhiko Katahira[2,1]　(1. United Graduate School of Agriculture , Iwate University(Japan), 2. Faculty of Agriculture, Yamagata University(Japan)）

10:45 AM - 11:00 AM

[6-1015-C-04] Application of Non-destructive Determination of Rice Amylose Content at Grain Elevators

*Edenio Olivares Diaz[1], Shuso Kawamura[1], Miki Matsuo[1], Toru Nagata[2], Shigenobu Koseki[1]　(1. Hokkaido University(Japan), 2. Hokkaido Research Organization Central Agricultural Experiment Station(Japan)）

11:00 AM - 11:15 AM

[6-1015-C-05] Cow Milk Progesterone Concentration Determination during Milking Using Near-infrared Spectroscopy

*Patricia Nneka Iweka[1], Shuso Kawamura[1], Tomohiro Mitani[2], Takashi Kawaguchi[3], Shigenobu Koseki[1]　(1. Hokkaido Univ.(Japan), 2. Field Sc. Center(Japan), 3. Orion Mach.(Japan)）

11:15 AM - 11:30 AM

Oral Session | Postharvest/Food Technology and Process Engineering

[6-1015-C] Postharvest/Food Technology and Process Engineering (6)

Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[6-1015-C-01] Spatially Resolved Interactance Spectroscopy to Estimate Degree of Red Coloration in Red-fleshed Apple Cultivar 'Kurenai-no-Yume'

*Xujun Ye[1], Sou Takada[1], Shuhuai Zhang[1] （1. Hirosaki University(Japan)）

Keywords: Red-fleshed apple, Kurenai-no-Yume, Red coloration, Interactance spectroscopy, Anthocyanin, Non-destructive estimation, Partial least squares regression

Reliable information about degree of red coloration in fruit flesh is essential for grading and sorting of red-fleshed apples. This study used spatially resolved interactance spectroscopy as a new rapid and non-destructive technique to estimate degree of red coloration in the flesh of a red-fleshed apple cultivar 'Kurenai-no-Yume'. A novel measurement device was developed to obtain spatially resolved interactance spectra at eight different light source-detector separations. A UV-Vis-NIR spectrometer connected to the detector fiber was used to obtain interactance spectra ranging from 190~1070 nm for 50 apple fruits. Anthocyanins in apple flesh were first extracted using a solvent extraction technique, and their contents were then quantified based on the absorbance measurements at 530, 620 and 650 nm obtained with a spectrophotometer. Partial least squares (PLS) regression analysis was performed to develop estimation models for anthocyanins content from spatially resolved interactance spectra. Results showed that the PLS models based on interactance spectra obtained at different light source-detector separations achieve different predictive accuracy for anthocyanins estimation. These results suggest that spatially resolved interactance spectroscopy could contribute to identifying an optimal light source-detector distance for implementing the interactance spectroscopy to assess the internal quality of red-fleshed apples. This new approach may be potentially applied to grading and sorting systems for red-fleshed apples in fruit industry.

Spatially Resolved Interactance Spectroscopy to Estimate Degree of Red Coloration in Red-fleshed Apple Cultivar 'Kurenai-no-Yume'

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ABSTRACT

Reliable information about degree of red coloration in fruit flesh is essential for grading and sorting of red-fleshed apples. This study used spatially resolved interactance spectroscopy as a new rapid and non-destructive technique to estimate degree of red coloration in the flesh of a red-fleshed apple cultivar 'Kurenai-no-Yume'. A novel measurement device was developed to obtain spatially resolved interactance spectra at eight different light source-detector separations. A UV-Vis-NIR spectrometer connected to the detector fiber was used to obtain interactance spectra ranging from 190~1070 nm for 50 apple fruits. Anthocyanins in apple flesh were first extracted using a solvent extraction technique, and their contents were then quantified based on the absorbance measurements at 530, 620 and 650 nm obtained with a spectrophotometer. Partial least squares (PLS) regression analysis was performed to develop estimation models for anthocyanins content from spatially resolved interactance spectra. Results showed that the PLS models based on interactance spectra obtained at different light source-detector separations achieve different predictive accuracy for anthocyanins estimation. These results suggest that spatially resolved interactance spectroscopy could contribute to identifying an optimal light source-detector distance for implementing the interactance spectroscopy to assess the internal quality of red-fleshed apples. This new approach may be potentially applied to grading and sorting systems for red-fleshed apples in fruit industry.

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1. INTRODUCTION

Red-fleshed apples are welcomed by consumers because of their unique flesh colors and additional health benefits offered by more anti-oxidants present in the flesh. 'Kurenai-no-Yume', literally called "crimson dream", is a new red-fleshed apple cultivar bred by Hirosaki University, Japan (Igarashi et al., 2010). Like many other red-fleshed apple varieties, 'Kurenai-no-Yume' suffers from significant difference in flesh red coloration among individual fruits. The degree of red coloration in the flesh cannot be known unless the fruit is cut. Such destructive method can be used to check fruit samples but cannot be applied to all fruits. And checking of only a limited number of samples cannot accurately reflect the degree of red coloration in other fruits even they are from the same batch. Therefore, there is a need to develop an accurate, rapid and non-destructive technique for determining the degree of red coloration for individual fruits. In our previous work, an interactance device was developed to collect interactance spectra for investigating red coloration in apple fruits (Ye et al., 2017). The device obtained interactance spectra with a constant distance between light source and detector fiber. Several models were developed for estimating the degree of red coloration in the flesh, with different levels of accuracy achieved. As a further step of this research, we recently developed a novel measurement device to obtain spatially resolved interactance spectra from apple fruits. The new device could obtain interactance spectra with a detector fiber, whose distance from light source could be adjusted to eight specified distances. In this study, we employed the new device to investigate the feasibility of spatially resolved interactance spectroscopy for estimating the degree of red coloration in the flesh of 'Kurenai-no-Yume' apples.

2. MATERIALS AND METHODS
2.1. Fruit material and sample preparation

'Kurenai-no-Yume', a red-fleshed apple cultivar, was used in this study. The fruit is sweet, mildly tart, rich in anthocyanin, and is delicious either fresh or cooked. The natural red color of flesh remains distinct even after cooking or processing, making it particularly useful for the creation of richly colored apple products (Hirosaki University, 2016). Therefore, it is welcomed by both consumers and manufacturers of apple fruit products.

Trees of this precious cultivar were cultivated in Fujisaki Farm of Hirosaki University. The fruits were harvested in a timely manner and carefully handled during harvest and transport. After screening them to ensure a high degree of fruit size uniformity, a total of 50 fruit samples were selected for the experiment. The flesh of fruit samples showed a large variation in the degree of red coloration (Fig. 1). Further, it was also found that the fruit skin color did not exactly reflect the color of the flesh, suggesting the difficulty to discriminate the flesh color simply based on the skin color of the fruit.

Figure 1. Different degree of red coloration in two 'Kurenai-no-Yume' apple fruits.

2.2. Spatially resolved interactance measurement system

A spatially resolved interactance measurement system was developed for collecting interactance spectra for apple fruits. The system is composed of a halogen light source generator LA-150ue-A (Hayashi Co., Japan), a ring illuminator (Hayashi Co., Japan) integrated with a self-made movable detector fiber, and a mini-spectroscope BLACK-Comet-SR100 (StellarNet Inc., USA) (Fig. 2a). During measurement, the ring illuminator was tightly placed on the fruit surface (Fig. 2b), allowing the halogen light to pass through the fruit skin and enter the flesh, and the amount of light that returned to the detector after scattering in the flesh was measured.

The mini spectroscope covers ultraviolet, visible and near infrared spectroscopic wavelengths from 190-1070 nm. The mini spectroscope is connected to a computer, and the interactance measurements for the sample are recorded when the system is operated by the software SpectraWiz (StellarNet Inc., USA) installed on a PC. Once the data is recorded, the interactance spectra for each measurement can be exported with the software for further analysis.

a). Experiment instruments   b). Measurement of interactance
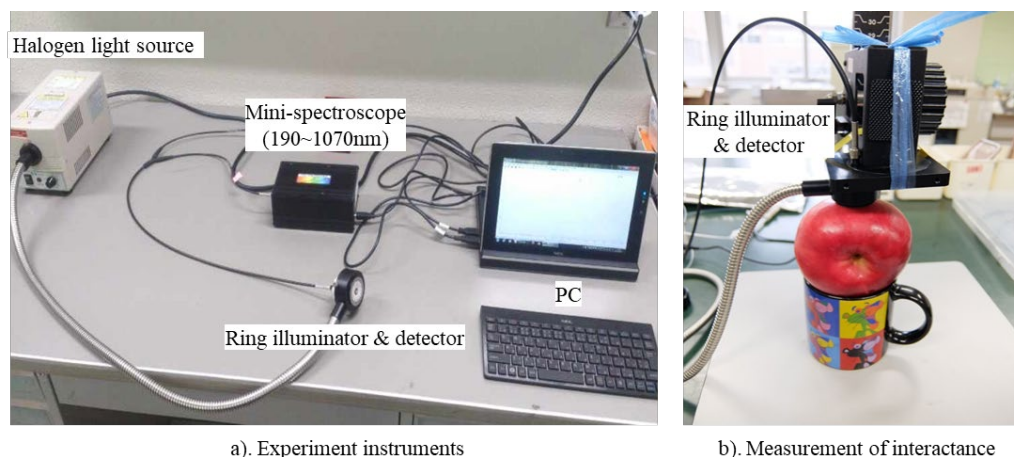
Figure 2. Instruments and experimental setup.

The working principle of the above system is described in more detail below. Figure 3a shows the structure of the ring illuminator integrated with the detector. The light from the halogen light source generator enters the ring illuminator through connecting fibers and forms a ring-type light beam (yellow ring) (Fig. 3b). The outer black cover and the inner light shield (black area) block the light that is directly reflected from fruit surface (Fig. 3b). The hole in the center serves as an entrance slit for the light that comes back to the detector after scattering in the flesh and passing back through the fruit skin (Fig. 3ab). In this structure, the light source (ring-type beam) and detector (entrance slit) are positioned parallel to each other, thus light due to specular reflection cannot directly enter the detector. The fibers leading to the source and detector are parallel to each other and in contact with the product (Fig. 2a). Furthermore, in this structure, the detector is designed to be movable rather than being fixed in the center point of the device. This enables the device to obtain spatially resolved interactance spectra from apple fruits. The new device could obtain interactance spectra with the detector fiber, whose distance from light source could be adjusted to eight specified distances. In Figure 3b, the detector fiber is placed in the center (label 8), which has the farthest distance between the light source and the detector, and the labels 1 to 15 represent different positions of the detector fiber at eight different light source-detector separations.

a). Ring illuminator and detector fiber.   b). Schematic diagram.
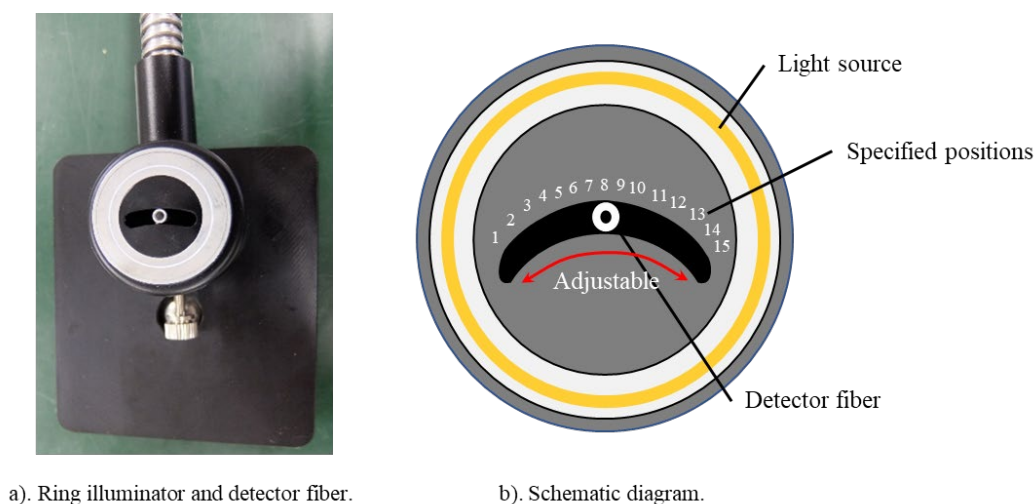
Figure 3. Ring illuminator and detector used to acquire interactance measurements.

2.3. Quantification of anthocyanins contents in apple flesh

In addition to the skin, anthocyanins are also present in large amounts in the flesh of red-fleshed apples. It can be increased up to several hundred-fold in red flesh compared with white flesh (Wang et al., 2014). Therefore, the anthocyanins content can be used as an indicator for the degree of red coloration in the flesh.

After the interactance measurements, the fruit skins of the measured positions were peeled, and the flesh under the peeled skins was cut separately. The cut flesh was weighed and ground to a pulp in a mortar using a pestle. The pulped flesh was subsequently extracted with 10 ml 1% ethyl acetic acid in methanol. The residue was re-extracted multiple times until all pigments were removed. The solvents containing anthocyanins were filtered through a filter paper and then transferred into a separating funnel. After the extractions, the anthocyanins absorbances at 530, 620 and 650 nm were measured with the Spectrophotometer U-2000S (Hitachi Co., Japan), based on which the anthocyanins contents in the flesh were calculated.

2.4. Data analysis and model development

The spatially resolved interactance spectra were used to relate to the anthocyanins content of the apple flesh. The partial least squares (PLS) regression, a standard calibration method for analyzing spectral data (Ramadan et al., 2004), was used to develop the prediction models. We used the R package 'pls' for modeling analysis (Mevik and Wehrens, 2007). In the modelling, we employed the leave-one-out

method for cross validation, in which potential models are calculated by excluding only one observation at a time (Kohavi, 1995).

3. RESULTS AND DISCUSSION
3.1Anthocyanins content in apple flesh
The anthocyanins content in the flesh ranged from 0.47 to 42.24 mg/g for the fruit samples, with an average of 9.19 mg/g and standard deviation of 8.95 mg/g, respectively, showing a tremendous difference in the anthocyanins content among fruits, which is responsible for the different degree of red coloration in the flesh among fruits.

3.2. Characteristics of spatially resolved interactance spectra
The spatially resolved interactance spectra were collected by the detector at eight different light source-detector separations. Because no or little interactance was detected in the short wavelength range, the interactances only for the wavelength range from 500 nm to 1070 nm were illustrated (Fig. 4). In Figure 4, except for label 8 of the specific positions (the center of the device), every two of other specific positions, such as labels 1 and 15, labels 2 and 14, etc., have the same distance from the light source, and therefore the averaged interactances of these paired positions were calculated.

The spatially resolved interactance spectra showed significant differences in the signal intensities of interactances among different specific positions, though all specific positions showed a similar pattern along the wavelength range (Fig. 4). The specific position label 8 (the center of the device) showed the lowest interactances, and the interactances increased gradually as the distances of the specific positions from the light source became closer (the spectra shifted upwards). Furthermore, the overall spatially resolved interactances of the fruit sample with a high anthocyanins content were lower than those of the fruit with a lower anthocyanins content, particularly within the 500-660 nm wavelength range. These results suggest that the spatially resolved interactances in these wavelength ranges may provide useful information about the anthocyanins content in apple flesh.

a). Fruit sample with high anthocyanins content   b). Fruit sample with low anthocyanins content
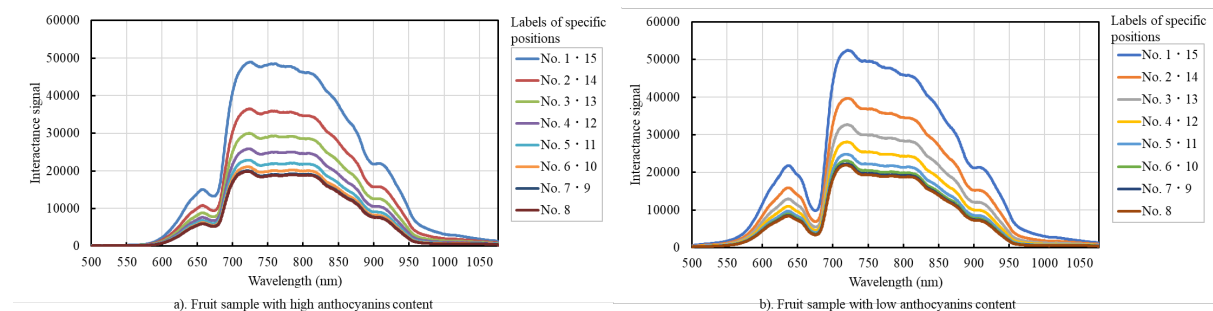
Figure 4. Characteristics of spatially resolved interactance spectra obtained for two fruit samples with high (a) and low (b) anthocyanins contests, respectively.

3.4. PLS models
The averaged interactances of the eight different light source-detector separations were used to develop PLS models. Figure 5 shows the predictive performance of the models for both the calibration and cross validation. It was found that the specific positions of labels 4 and 12 achieved the best predictive performance in both the calibration and cross validation, and the light source-detector separations (labels 1 and 15, 2 and 14, and 3 and 13) with a shorter distance showed a significantly lower predictive accuracy. This might be attributed to the fact that more interactions occur between scattered light and fruit tissues when light passes through a longer distance within the flesh of a fruit, and these interactions enable the collection of interactance spectra with more information about the internal properties of fruits. Nevertheless, the central position (label 8), which has the longest light source-detector separation, demonstrated the lowest prediction accuracy in cross validation, though its performance was better than its neighboring measurement positions. This suggests that when the light source-detector distance exceeds a certain distance limit, the interactance that can be detected might become much less informative because most of the light is absorbed by the flesh and thus cannot reach the detector.
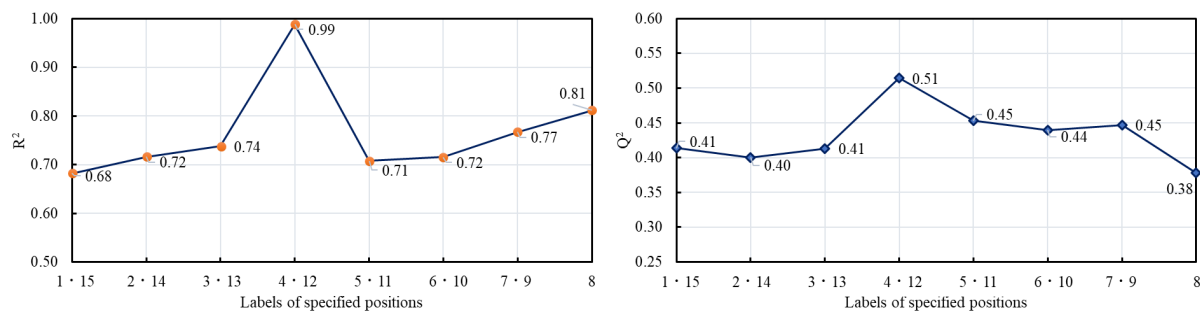
Figure 5. Coefficients of determination for calibration (R^2) and cross validation (Q^2) of PLS regression analysis based on averaged interactances obtained at eight different light source-detector separations.

The PLS model based on the averaged interactances for the optimal light source-detector separations (specific positions label 4 and 12) identified in the previous analysis was developed. Scatter plots of the estimated against the measured anthocyanins contents and the coefficients of determination for the predictions are illustrated in Figure 6. The model achieved a good predictive performance in the calibration ($R^2=0.99$), and a reasonable predictive accuracy ($Q^2=0.51$) was also obtained. This result demonstrated the possibility of estimating the anthocyanins content in apple flesh from the interactance spectra obtained with a non-destructive approach.
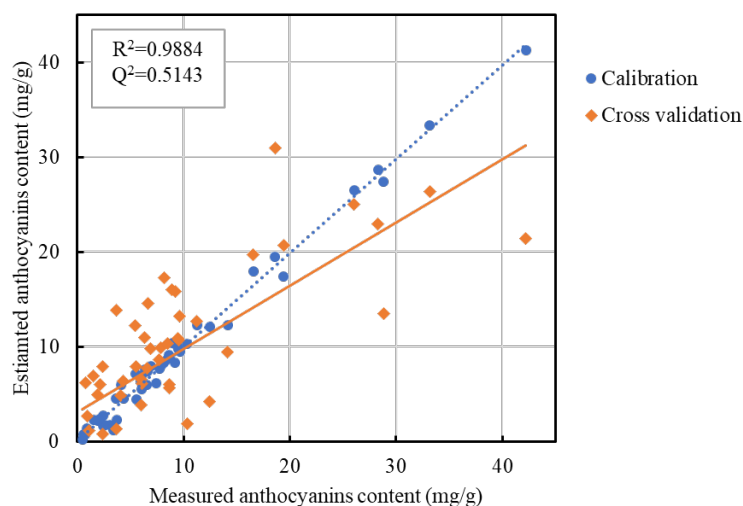
Figure 6. Predictive performance of the PLS model based on averaged interactances obtained at the optimal light source-detector separations (specific positions labels 4 and 12).

4. CONCLUSION

This study investigated the possibility of spatially resolved interactance spectroscopy to estimate the degree of red coloration in the flesh of a red-fleshed apple cultivar 'Kurenai-no-Yume'. A novel measurement device was developed to obtain spatially resolved interactance spectra at eight different light source-detector separations. A UV-Vis-NIR spectrometer (190~1070 nm) connected to the detector fiber was used to collect the spatially resolved interactance spectra for apple fruits. Anthocyanins contents in apple fleshes were quantified using a spectrophotometric method. Partial least squares (PLS) regression analysis was performed to develop predictive models for anthocyanins content from spatially resolved interactance spectra. Results showed that the PLS models based on interactance spectra obtained at different light source-detector separations achieve different predictive accuracy for anthocyanins estimation and increasing the detector's distance from light source tends to

improve the predictive accuracy. This might be attributed to the fact that more interactions occur between scattered light and fruit tissues when incident light passes through a longer distance within the flesh of a fruit, and these interactions enable the collection of interactance spectra with more information about the internal properties of fruits. However, a further increase in the light source-detector distance would lead to reduced predictive accuracy, as the interactances become less informative because most of the light is absorbed by the flesh and thus cannot reach the detector to be captured. This study suggests that spatially resolved interactance spectroscopy could contribute to identifying an optimal light source-detector distance for implementing the interactance spectroscopy to assess the internal quality of red-fleshed apples. This new approach may be potentially applied to grading and sorting systems for red-fleshed apples in fruit industry.

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Oral Session | Postharvest/Food Technology and Process Engineering
[6-1015-C] Postharvest/Food Technology and Process Engineering (6)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[6-1015-C-02] Use of hyperspectral imaging to separate cultivars and evaluate the internal quality of nectarines

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Keywords: Stone fruit, Hyperspectral transmittance imaging, Ripeness monitoring, Cultivar discrimination, Internal quality, Computer vision

Visible–near-infrared hyperspectral imaging (450-1040 nm) was studied in reflectance and transmittance modes to assess the internal physicochemical properties and sensory perception of ‘Big Top’ and ‘Magique’ nectarines (*Prunus persica* L. Batsch var. *nucipersica*) (yellow and white-flesh cultivar, respectively) during ripening. These properties were successfully correlated to the Ripening Index (RPI) and the Internal Quality Index (IQI). During ripeness under controlled conditions, hyperspectral images of the intact fruits were acquired and their physicochemical properties (flesh firmness, total soluble solids, titratable acidity and flesh colour) were analysed. Moreover, a model to discriminate between both cultivars were developed. IQI and RPI were correlated to the hyperspectral images by using Partial Least Square (PLS) regression with proper variables selection. Optimal results were obtained with R^2 (and RPD) values of 0.89 (2.7), 0.90 (3.1), 0.90 (2.8) and 0.88 (2.7) for RPI and IQI in ‘Big Top’ and ‘Magique’ nectarines, respectively.

In addition, the emergence of new cultivars in the market with similar appearance but different sensory properties can cause confusion among the consumers, being necessary the development of new tools capable of discriminating these cultivars in an automated and non-destructive way. PLS-DA was used to obtain the best classification model to distinguish intact fruits of both cultivars using individual pixel spectrum and mean spectrum of each fruit, and then projecting the model onto the complete surface of fruits in a validation or prediction set. The results indicated that mean spectrum approach was the most accurate, 84.4 % vs. 94.4 %. Moreover, a comprehensive wavelength selection was performed, reducing the dimensionality of the hyperspectral images using the regression coefficients of the mean spectrum PLS-DA model, obtaining an accuracy of 96.3 % by using 14 optimal wavelengths.

A PLS model of IQI prediction was used to transfer the calibrated results to each pixel of the image and to visualise the evolution of ripeness on the surface of the fruits, and also to represent the probability of whether any pixels belongs to one or another cultivar.

Finally, the internal quality of the nectarines was inspected using hyperspectral transmittance imaging during their ripening under controlled conditions. The detection of split pit disorder and classification according to an established firmness threshold were performed using PLS-DA. The prediction of the IQI related to ripeness was performed using PLS-R. The most important variables were selected using interval-PLS. As a result, an accuracy of 94.7 % was obtained in the detection of fruits with split pit of the ‘Big Top’ cultivar. Accuracies of 95.7 % and 94.6 % were achieved in the classification of the ‘Big Top’ and ‘Magique’ cultivars, respectively, according to the firmness threshold. The internal quality was predicted through the IQI with R^2 values of 0.88 and 0.86 for the two cultivars. The results obtained indicate the great potential of hyperspectral

Use of hyperspectral imaging to separate cultivars and evaluate the internal quality of nectarines

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[6-1015-C] Postharvest/Food Technology and Process Engineering (6)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[6-1015-C-03] Evaluating the Performance of Unmanned Crop Sensing Robot for Rice

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Keywords: Crop Sensing, Unmanned Ground Vehicle (UGV), Precision Agriculture, Rice

Precision Agriculture has emerged as a new scientific field that seeks to drive agricultural productivity while minimizing its environmental impacts. As the demand for food increases, farmers are in search of technology that would allow them cultivate more land with less labour at the same time increasing their productivity. In rice cultivation, this has led to the adoption of technologies such as Unmanned aerial vehicles (UAV) for crop monitoring. While this has increased precision from traditional satellite images, it still has the limitation of being restricted to capturing images of the crop canopy. Unmanned Ground Vehicles (UGV) on the other hand has the potential to capture a wider range of data with pin point accuracy. This paper reports on the work done thus far in evaluating the performance of a field robot developed by the World Wide Food Platform, Japan for rice crop sensing. The study was conducted in 3 rice fields at the Yamagata University's Farm in Takasaka, Tsurouka, Japan and a Farmers' Field in Mikawa, Yamagata, Japan. The cultivation system in the fields were transplanting, hilldrop and broadcasting at Takasaka, while in Mikawa drill seeding was done. The robot is equipped with sensors for temperature, humidity, sunlight, wind speed, soil temperature, water level and temperature and cameras (Sony FDR-X3000) for image capture. RTK GPS was used for location logging with an accuracy of 5 cm. Data captured were mapped into QGIS 3.4 for visualization an analysis of growth parameters every two weeks after germination, with observations made on the robots' maneuverability in the various field conditions. Plant height, leaf and tiller number, and SPAD values were collected manually in each field to compare for image data. It was found that the robot was able to maneuver in different field conditions without major issue, utilizing the reverse function instead of turning full circle appears to be the most efficient method for turning while causing minimal damage to young seedlings. The weight distribution will have to be considered to obtain optimum performance in deep fields. Data collected from the array of sensors and cameras provides location specific information throughout the field and can be used to guide farmers in precision management.

Evaluating The Performance of Unmanned Crop Sensing Robot for Rice

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ABSTRACT

Precision Agriculture has emerged as a new scientific field that seeks to drive agricultural productivity while minimizing its environmental impacts. As the demand for food increases, farmers are in search of technology that would allow them cultivate more land with less labour at the same time increasing their productivity. In rice cultivation, this has led to the adoption of technologies such as Unmanned aerial vehicles (UAV) for crop monitoring. While this has increased precision from traditional satellite images, it still has the limitation of being restricted to capturing images of the crop canopy. Unmanned Ground Vehicles (UGV) on the other hand has the potential to capture a wider range of data with pin point accuracy. This paper reports on the work done thus far in evaluating the performance of a field robot developed by the World Wide Food Platform, Japan for rice crop sensing. The study was conducted in 3 rice fields at the Yamagata University's Farm in Takasaka, Tsurouka, Japan and a Farmers' Field in Mikawa, Yamagata, Japan. The cultivation system in the fields were transplanting, hilldrop and broadcasting at Takasaka, while in Mikawa drill seeding was done. The robot is equipped with sensors for temperature, humidity, sunlight, wind speed, soil temperature, water level and temperature and cameras (Sony FDR-X3000) for image capture. RTK GPS was used for location logging with an accuracy of 5 cm. Data captured were mapped into QGIS 3.4 for visualization an analysis of growth parameters every two weeks after germination, with observations made on the robots' maneuverability in the various field conditions. Plant height, leaf and tiller number, and SPAD values were collected manually in each field to compare for image data. It was found that the robot was able to maneuver in different field conditions without major issue, utilizing the reverse function instead of turning full circle appears to be the most efficient method for turning while causing minimal damage to young seedlings. The weight distribution will have to be considered to obtain optimum performance in deep fields. Data collected from the array of sensors and cameras provides location specific information throughout the field and can be used to guide farmers in precision management.

Keywords: Crop Sensing, Unmanned Ground Vehicle (UGV), Precision Agriculture, Rice

1. INTRODUCTION

Agriculture plays a critical role in feeding the 7.6 billion people in the world. With the available labour for agriculture declining as demand for food increases, farmers and researchers are exploring the application of technologies that will allow for more land to be cultivated with a reduced labour force while at the same time maintaining productivity. In rice cultivation, this has led to the adoption of technologies such as Unmanned aerial vehicles (UAV) for crop monitoring. While this has increased precision from traditional satellite images, it still has the limitation of being restricted to capturing images of the crop canopy. Unmanned Ground Vehicles (UGV) on the other hand has the potential to capture a wider range of data with pin point accuracy. Work on unmanned vehicles with respect to rice has mainly been focused on adapting conventional farm machinery to execute task without human intervention. Commercially available

tractors can be modified into autonomous vehicles by adding the electronics and communication devices necessary for autonomous operation in agricultural fields (Aravind et al. 2017). TAKAI et al. (2010) modified a crawler-type tractor and evaluated its accuracy for autonomous navigation using RTK-GPS and IMU navigation sensors while Tamaki et al. (2013) explored a robot system for tillage, transplanting and harvesting rice by adapting conventional machinery. They showed it was possible for one operator to operate more than one machine thereby reducing the time and labour necessary for a specific task. Because of their size and potential injury to crop, the adapted farm machinery is not suitable for crop sensing, as a result, this aspect of crop production is being done by UAVs, stationary infield sensors and human labour. Compared to UAVs and stationary sensors, a ground robot designed to traverse the rice field with minimal damage to crop offers more detail crop sensing information with greater accuracy. In this study, we evaluate the performance of a field robot developed by the World Wide Food Platform, Japan for rice crop sensing.

2. MATERIALS AND METHODS
2.1 Location

The study is being conducted in three experiment plots for rice at the Yamagata University's Farm in Takasaka, Tsuruoka, Japan and one farmer field in Mikawa, Yamagata, Japan. The cultivation system in the fields were transplanting, hilldrop and broadcasting at Takasaka while drill seeding was done at Mikawa. Field dimensions are 30m x 8m for the transplant and hill drop fields while the drill seeding and broadcast fields were 100m x 30m.

2.2 Equipment

2.2.1 Field Robot outline

The field robot used in this study was developed by the World Wide Food Platform, Japan which is a consortium of Tech companies, Universities and Farmers' Organizations. It is 140 cm in length, 120cm width and 145 cm height. Approximate weight is 180 kg. Two 12 volt batteries supply power to 4 brushless electric motors (200W, 3000r/min Orientalmotor, Model BLVM620KM-GFS, Japan) that rotate each the four wheels. The wheel diameter is 65cm and is of similar type used on rice transplanting machines. The drive controller is Ardupilot pixhawk 2 while the operation of the robot is by remote control.

Figure 1. Front & Side view of crop sensing robot and remote controller

2.2.2 Sensor Box
A sensor box containing sensors for Temperature and Humidity (SHT31 module), sunshine (Sanko PV Array Pryanometer PVSS-01), Airspeed (Powerday Airspeed sensor), water temperature (Thermistor 103AT-11), water level (Distance sensor MB1242) is mounted unto the robot with RTK GPS attached. Data recorded by sensorbox is stored on micro SD card.

2.2.3   RTK GPS

Here+ RTK GPS rover and base station was used with Mission Planner for GPS logging. Base station data was recorded for 5 days to obtain an accuracy of 5cm for each plot.

2.2.4   Camera

Two Sony FDR-X3000 action cameras were used for image capture. Images were captured in HD (1920 x 1080 pixels). Three orientations were explored with mounting cameras on the robot with the aim of acquiring suitable images. In the first instance Fig.2 (a) the camera was mounted directly on the robot facing downward at an approximate angle of 45 degrees. In the second orientation, camera mounts were used to extend the cameras 25cm from the robot with an approximate angle of 45 degrees, while the third orientation consisted extending the cameras 90 cm from the robot at an angle of 90 degrees. The images were captured from a height of 110 cm with the distance between the two cameras 100 cm. Considerations for suitable images were those that were clear, presented a full picture of the plants without any obstruction or unnecessary scenery that would necessitate further processing or transformation before image analysis can be executed. As such, the camera orientation was adjusted as described above until the desired results were achieved.

(a)   (b)   (c)
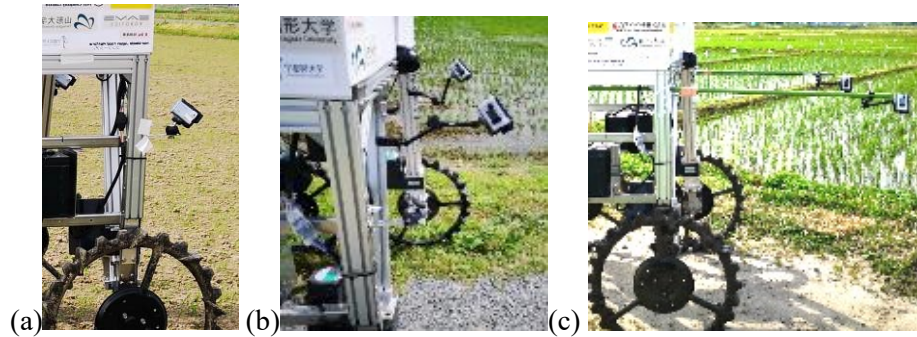
Figure 2. Camera orientation on Robot

2.3 Data collection

2.3.1 Data Collection with Robot

Data collection with the robot commenced at the end of May 2019 when plants were at the 5 leaf stage. In addition to image and sensing data, observations were made on robot maneuverability in rice fields, damage to plants, operation time and speed. Figure 3 shows the base station set up for data collection. The RTK antenna is placed at the same location where base station data was captured for 5 days to obtain an accuracy level of 5cm. It is connected to the RTK base station which is connected to the Note PC via USB. The link between the PC and robot is made via a UDP WiFi connection with pixhwak in the sensor box through the Misson Planner Software.
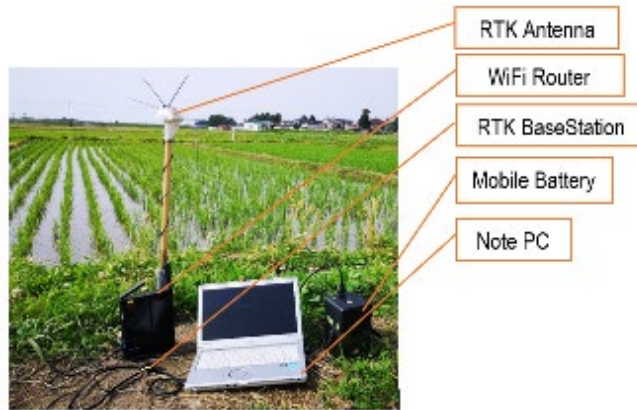
Figure 3. Base Station set up during data Collection

2.3.2 Manual Data collection

Growth data was collected manually either directly after data collection with robot or within 48hours depending on weather conditions. Three test plots were randomly selected in each field and demarcated. SPAD, plant height, number of tillers, and leaf number surveyed at two week intervals after germination. SPAD was measured using SPAD-502 Plus (Konica Minolta, Japan).

2.4 Data Processing

Data recorded by sensor box were converted from .bin to .log files in using Mission Planner (Version 1.3 by Michael Osborne). A user defined Python (3.6) script was then use to parse data from the .log files and write to .csv file. The .csv file was then imported into QGIS (version 3.4 LTR) and vector layers created for areas of interest; Altitude, Temperature, Humidity, Air Speed, Sunshine. Images captured were imported into QGIS via the Import Photos plugin to provide an image for each point captured.

3. RESULTS AND DISCUSSION
3.1 Robot

3.1.1 Maneuverability in Rice Fields.

Observations were made on the robot's ability to maneuver in the rice fields. The robot covers four rows in one pass with wheels travelling between rows. It is able to effect turns by stopping motors on one side while the other side continues to move (like pivot system). The smallest turning diameter on dry surface was 160 cm, however in effecting this turn there is sliding /dragging of the unmoving wheels of approximately 30 cm. Attempting such turn in flooded field conditions resulted in longer sliding distance and dislodged or damaged seedlings. In some cases, the wheel also locked and a reboot was required to free the wheels.

Executing a gentler turn requires a turning diameter of approximately 250 cm. In this turn all wheels move turning slightly until the turn is complete. This method reduces the sliding/dragging of wheel and results in very few damaged or dislodge seedlings. Executing turns in this manner however requires more space and also will leave gaps in the field at turning points with respect to data collection (Fig. 4 a) before the robot can enter the set of rows.

The best turning method devised thus far involves utilizing the robot's ability to reverse freely. In this method, instead of turning 360 degrees, just prior to reaching the end of the row, the robot makes a slight turn to exit the row and then reverse into the new set of rows (Fig.4b), only when it reaches the end of those rows, it turns slightly to enter the new set of rows by going forward. Operation using this 'switchback' method reduces the amount of degree the robot has to turn resulting in minimal damage or dislodgement of seedlings. It also makes it easy to enter the next set or rows without missing any data. Figure 4(c, d) shows the actual path travelled by the robot using the two methods.
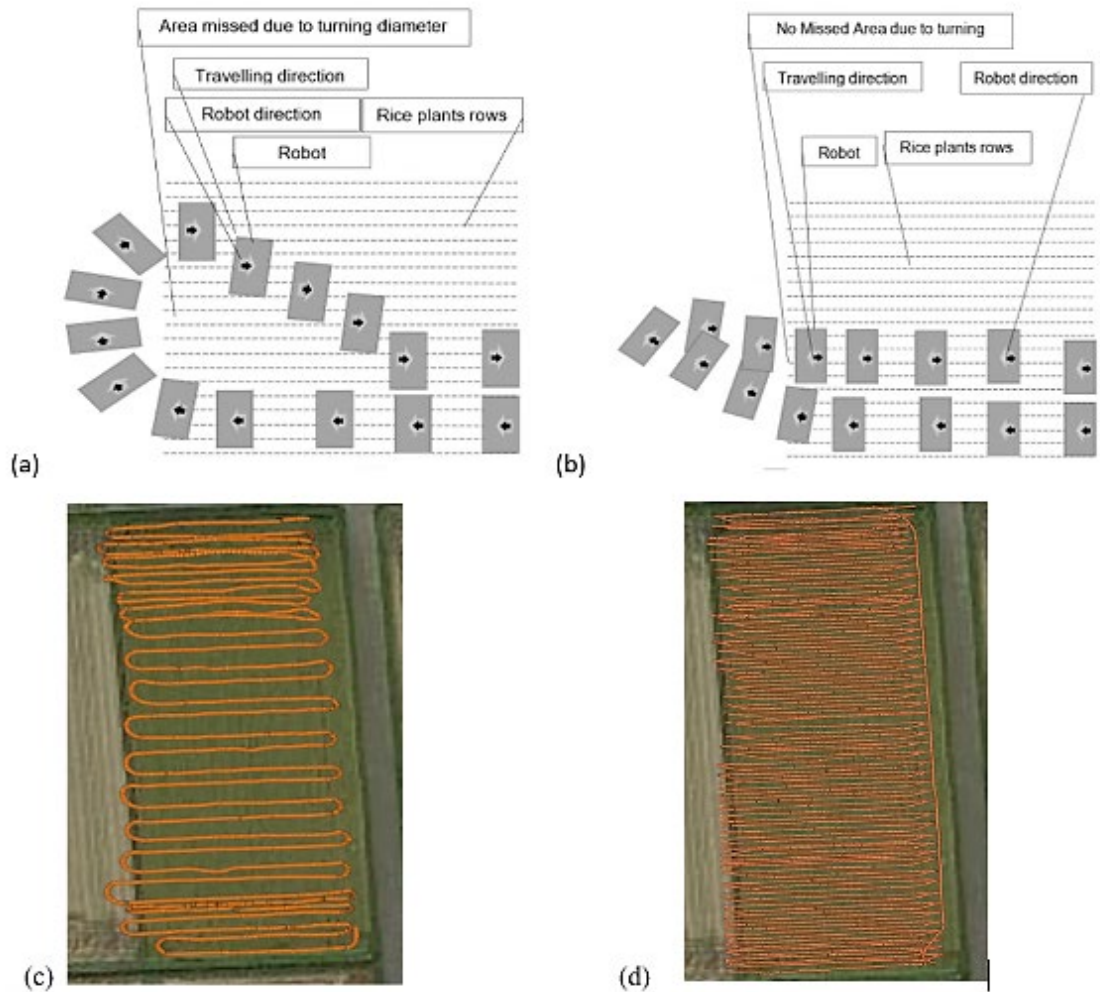
Figure 4. Turning methods (a,b) and actual path travelled by robot using each method (c,d)

3.1.2 Speed

Speed of robot was recorded by flight controller in meters per second (m/s) which ranged from 0.1 m/s at its lowest speed to 0.6 m/s with 0.3-0.4 m/s being the most common /steady speed. Time require to survey 0.3 ha (30m x 100m) field ranges between 2.5-3 hours depending on the amount of overlap (rows) is required.

3.1.3 Field Condition

The robot was able to traverse various rice fields in flooded condition without any major issue. The wheel reduces its ability of getting stuck. It was able to traverse in a field where the mud was approximately 35cm in depth, however very wide turns had to be taken in this situation. Attempts to make small or sharp turns resulted in churning of mud and locking of wheels. It was able to operate freely when going straight and turn by making wide gentle turns. One area that may require further consideration is the weight distribution as there is a tendency to tilt forward or backward especially in deep fields when torque is applied (Fig.5).

Figure 5. Robot in Deep Field Condition

In fields in which the water was drained and the mud had stiffened to some extent, it was observed that the robot tilted heavily rearward while the front wheels tend to lift off the ground when torque was applied with the threat of tilting over (Fig. 6).

Figure 6. Robot in drained field

This situation arose due to the lack of water which cause the mud loosened by the front wheels to stick to the back wheel adding more weight and friction with the soil thus requiring more torque for it to move. The front wheels are not affected by such impediments as such, when the power is applied it surges forward while the rear is being held back resulting the front wheels going aerial. It is suspected that this situation may be remedied by extending the wheel based of the robot and adding scrapers to the wheels to remove excess mud that may become stuck on it while operating in drier fields.

3.2 Camera Orientation

Images captured on the 30th May, 10th June and 2nd July 2019 in Mikawa field from the three camera orientation is shown in Fig3. The first camera orientation (Fig.7a) captures the surrounding scenery in the image in addition to the plants, this is reduced to some extent in images captured in camera orientation

two (Fig.7b) however the angle obscures details in parts of the image. Adjusting these mounting methods to orient the camera at 90 degrees results in images that include the wheel and parts of the robot. Camera orientation three (Fig.7c) that extends from the robot and captures the images from directly above appears to be the most suitable for image capture as no unnecessary information is contained in the images and details are not obscured in any part of the image.

(a)   (b)   (c)

Figure 7. Images captured by different camera orientations

3.3 Data

The data collected by the robot are field gives information of the field environment, record measurements and captures images. Data collected for field environment includes temperature, humidity, sunshine and airspeed. While these conditions may vary depending on climatic conditions, time of day data is collected as well as over the total time taken to collect the data, as more data is collected patterns may emerge that point to specific location based issues in the field irrespective of the factors mentioned above. In Figure 7 data collected by the robot on the 2nd of July 2019 is shown as QGIS layers for temperature, humidity, sunshine and airspeed for the Mikawa field. The temperature layer (Fig.7a) shows how temperature varies across the field with the highest temperature being observed around the center and reduces as it gets to the lower end. While this may be due to the time data collection started and conditions changing in the duration it take for the robot to reach the lower end of the field. If a similar pattern persist as more data is collected at different time and prevailing weather conditions, it give some insight as to what is happening in the field with respect to temperature. Optimal Temperature for rice growth is considered to be 22-28°C, as such any increase in mean temperature or episodes of high temperature during sensitive stages of the crop may adversely affect the growth and yield of the crop (Krishnan, Ramakrishnan, Reddy, & Reddy, 2011).

(a)   (b)
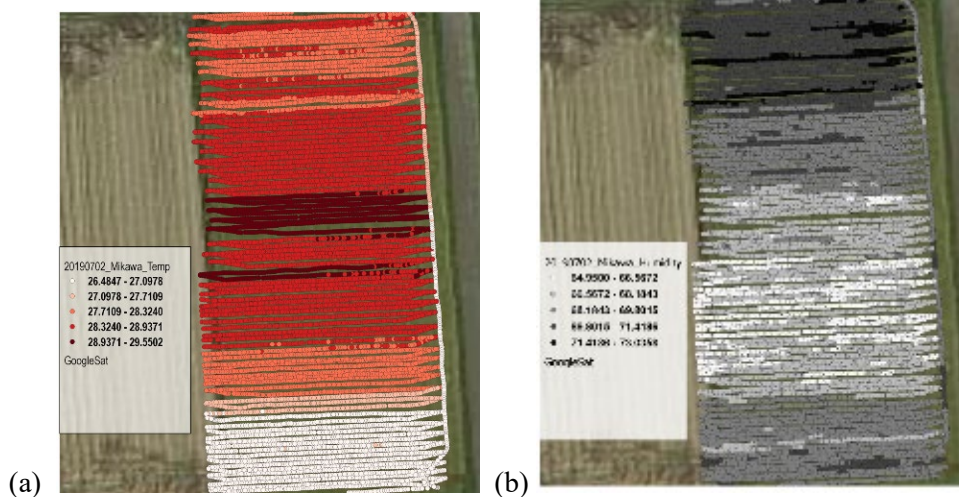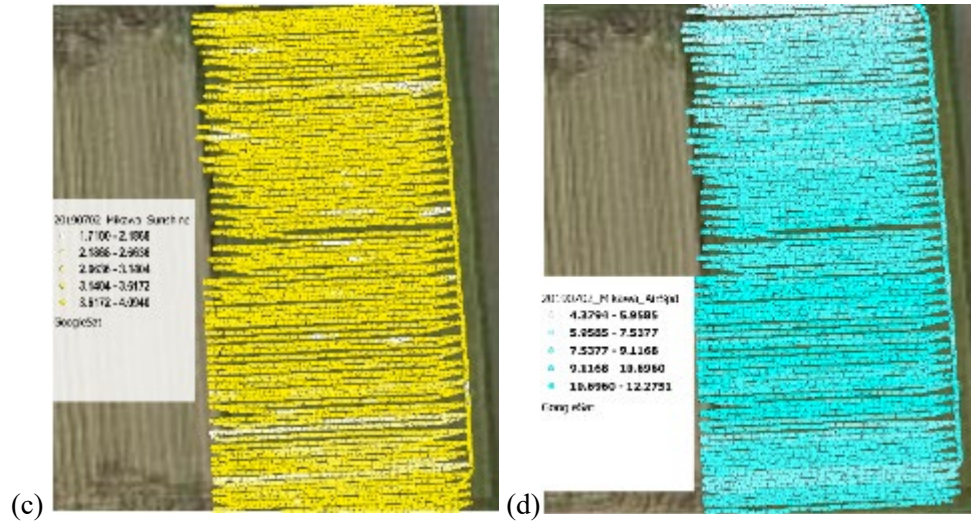
Figure 8. Data collected by Robot in Mikawa field, 2019/07/02: (a) Temperature, (b) Humidity, (c) Sunshine, (d) Airspeed.

The mean relative humidity during rice cultivation is generally negatively associated with solar radiation. Krishnan et al., (2011) discusses works by Morokuma and Yasuda,(2004), Nishiyama and Satake (1981) and Matsui et al., (1997) that reports on increases in spikelet sterility with increases humidity at high air temperatures. They further suggest that the effects of temperature on rice may be intermingled with those of relative humidity and solar radiation. Given the various effects these environmental factors may have on the crop, being able to monitor their distribution over the field may provide insights on how to better manage the field.

The altitude recorded by pixhawk indicates the robot's altitude in meters (m) from sea level. From the plot of this layer in QGIS, it is possible to infer how level the field is, and identify areas that appear too high or low. Figure 9 shows the plot of Altitude data collected at Mikawa on the 2nd of June 2018 where varying altitude is recorded over the field indicating that specific parts of the field is higher and lower than others.
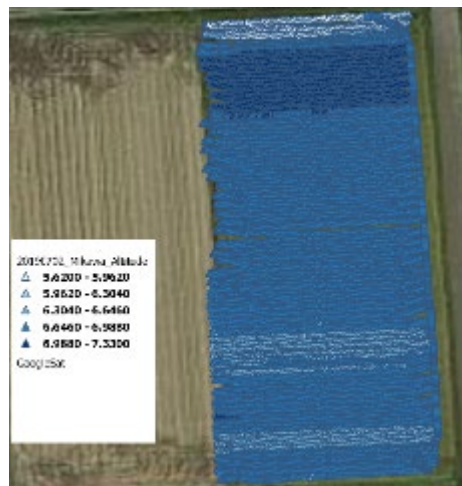
Figure 9. Plot of Altitude recorded in Mikawa Field on 2nd July, 2019

Image Data

Two Sony FDR-X3000 action cameras were used for image capture. Time lapse images at one image per second were captured in HD (1920 x 1080 pixels). The images were geotagged and imported into QGIS to provide an image for each point in the field. These images provide a clear picture at each point of the field with a level of detail not possible with UAVs without the use of expensive high specification cameras. From the images it was possible to identify the presence of weeds in the field as seed in Figure 10 which shows a sample image captured by the cameras. Future work will involve building deep learning AI to detect weeds, insect pests and disease in the field and generate location map for precision management. Attempts will also be made to extract growth data from images by analysis plant canopy for height, tiller and greenness.

Figure 10. Sample Image from Mikawa Field on 2nd July, 2019

4. CONCLUSION

The paper reports on the initial results obtained while evaluating the performance of crop sensing robot for rice. It was found that the robot was able to maneuver in different field conditions without major issues and utilizing the reverse function instead of turning full circle appears to be the most efficient method for turning while causing minimal damage to young seedlings. The weight distribution will have to be considered to obtain optimum performance in deep fields. Data collected from the array of sensors and cameras provides location specific information throughout the field and can be used to guide farmers in precision management. Given that this is the first season of testing, various applications and analysis will be explored with the data gathered to allow for refining and optimizing of the methodologies for next season.

ACKNOWLEDGMENT

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Oral Session | Postharvest/Food Technology and Process Engineering
[6-1015-C] Postharvest/Food Technology and Process Engineering (6)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[6-1015-C-04] Application of Non-destructive Determination of Rice Amylose Content at Grain Elevators

*Edenio Olivares Diaz[1], Shuso Kawamura[1], Miki Matsuo[1], Toru Nagata[2], Shigenobu Koseki[1] (1. Hokkaido University(Japan), 2. Hokkaido Research Organization Central Agricultural Experiment Station(Japan))
Keywords: Oryza sativa L., Amylose content, Near-infrared spectroscopy, Chemometric analyses, Quality, Practical application

Rice (*Oryza sativa* L.) is the most important staple food for people in a large part of the world. Starch, moisture, and protein are the major constituents comprising the rice endosperm. However, amylose content (AC), which is the percentage of amylose relative to total starch in the rice kernel, contributes to the texture and quality of cooked rice. Iodine-binding, also known as iodine colorimetry or amylose-iodine, is the only validated and most commonly used method for determining AC. But it is labor-intensive, time-consuming, chemical-dependent, and vulnerable to random error. Therefore, it is unsuitable for laboratory and/or industrial uses where large volumes of samples need to be processed. To overcome this shortcoming, near-infrared (NIR) spectroscopy in combination with chemometric techniques represents an alternative, validated method for assessing rice AC. In this study, we developed an accurate model for the non-destructive determination of AC at grain elevators. A dual-step calibration model was developed using data from 936 samples of 10 varieties of rice produced between 2008 and 2018 in various regions of Hokkaido, Japan. The collected rough rice samples were dried to approximately 15% w.b. of moisture content. Next, each dried sample was hulled to obtain brown rice. Finally, each brown rice sample was milled to 90.5 ± 0.2% of milling degree. Milled rice AC reference values (AC_{ref}), transmittance NIR spectra, and physicochemical properties were combined to develop the dual-step calibration model. The raw NIR transmittance spectra was preprocessed by applying a 2nd order Savitzky-Golay derivative with a 2nd polynomial order. Later, AC_{ref} values, transformed NIR spectra, and physicochemical properties were analyzed by partial least squares regression (PLS) and multiple linear regression (MLR) to develop an accurate dual-step calibration model. Our dual-step calibration model described low and ordinary amylose varieties models, which were developed individually. Both the low and ordinary amylose varieties models were calibrated using 2008-2017 production year samples (n = 903) and validated by distinct 2018 production year samples (n = 33), which were collected at a rice grain elevator. Next, the dual-step calibration model was created by merging the validation results of the low and ordinary amylose varieties models. Results indicated that AC_{ref} values were determined with high accuracy based on the low average standard error of the laboratory method (SEL) = 0.17% among varieties and production years. Also, the regression coefficients of each wavelength related to AC_{ref} for the optimal PLS factor indicated that the wavelength at 916 nm reported the highest spectral variation and thus correlated the most to AC. Moreover, validation statistics such as standard error of prediction (SEP) = 0.33% and ratio of performance deviation (RPD) = 5.09 indicated the high robustness and accuracy of the dual-step model, enabling more precise, accurate, and efficient rice quality screening at Japanese grain elevators.

Application of Non-destructive Determination of Rice Amylose Content at Grain Elevators

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ABSTRACT

Rice (*Oryza sativa* L.) is the most important staple food for people in a large part of the world. Starch, moisture, and protein are the major constituents comprising the rice endosperm. However, amylose content (AC), which is the percentage of amylose relative to total starch in the rice kernel, contributes to the texture and quality of cooked rice. Iodine-binding, also known as iodine colorimetry or amylose-iodine, is the only validated and most commonly used method for determining AC. But it is labor-intensive, time-consuming, chemical-dependent, and vulnerable to random error. Therefore, it is unsuitable for laboratory and/or industrial uses where large volumes of samples need to be processed. To overcome this shortcoming, near-infrared (NIR) spectroscopy in combination with chemometric techniques represents an alternative, validated method for assessing rice AC. In this study, we developed an accurate model for the non-destructive determination of AC at grain elevators. A dual-step calibration model was developed using data from 936 samples of 10 varieties of rice produced between 2008 and 2018 in various regions of Hokkaido, Japan. The collected rough rice samples were dried to approximately 15% w.b. of moisture content. Next, each dried sample was hulled to obtain brown rice. Finally, each brown rice sample was milled to $90.5 \pm 0.2\%$ of milling degree. Milled rice AC reference values (AC_{ref}), transmittance NIR spectra, and physicochemical properties were combined to develop the dual-step calibration model. The raw NIR transmittance spectra was preprocessed by applying a 2nd order Savitzky-Golay derivative with a 2nd polynomial order. Later, AC_{ref} values, transformed NIR spectra, and physicochemical properties were analyzed by partial least squares regression (PLS) and multiple linear regression (MLR) to develop an accurate dual-step calibration model. Our dual-step calibration model described low and ordinary amylose varieties models, which were developed individually. Both the low and ordinary amylose varieties models were calibrated using 2008-2017 production year samples (n = 903) and validated by distinct 2018 production year samples (n = 33), which were collected at a rice grain elevator. Next, the dual-step calibration model was created by merging the validation results of the low and ordinary amylose varieties models. Results indicated that AC_{ref} values were determined with high accuracy based on the low average standard error of the laboratory method (SEL) = 0.17% among varieties and production years. Also, the regression coefficients of each wavelength related to AC_{ref} for the optimal PLS factor indicated that the wavelength at 916 nm reported the highest spectral variation and thus correlated the most to AC. Moreover, validation statistics such as standard error of prediction (SEP) = 0.33% and ratio of performance deviation (RPD) = 5.09 indicated the high robustness and accuracy of the dual-step model, enabling more precise, accurate, and efficient rice quality screening at Japanese grain elevators.

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Oral Session | Postharvest/Food Technology and Process Engineering
[6-1015-C] Postharvest/Food Technology and Process Engineering (6)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room C (3rd room)

[6-1015-C-05] Cow Milk Progesterone Concentration Determination during Milking Using Near-infrared Spectroscopy

*Patricia Nneka Iweka[1], Shuso Kawamura[1], Tomohiro Mitani[2], Takashi Kawaguchi[3], Shigenobu Koseki[1] （1. Hokkaido Univ.(Japan), 2. Field Sc. Center(Japan), 3. Orion Mach.(Japan)）

Keywords: Bovine milk, Progesterone, Estrus status, Pregnancy diagnosis, Trend monitoring, Near-infrared spectroscopy, Automatic milking system

In the current dairy industry, an intensive demand for estrus detection and early diagnosis of pregnancy has been increasing. Progesterone is a steroid hormone that is secreted from corpus luteum into bovine blood and milk, and has a role of maintenance of estrus cycle and pregnancy. Therefore, progesterone concentration in cow milk is used as an important indicator of estrus detection and early diagnosis of pregnancy. Current method for milk progesterone determination requires a hormone extraction procedure that is time consuming, various types of instruments, reagents management, and various assay methods that are destructive in nature. In contrast, near-infrared spectroscopy (NIRS) is a time saving and non-destructive analytical method that can be used for online real-time determination of milk constituents content such as milk fat, protein, lactose, milk urea nitrogen and somatic cell count. However, there has been limited study on using NIRS for online real-time determination of progesterone concentration in milk during milking. Thus, the objective of this study was to develop an online real-time NIR spectroscopic sensing system for milk progesterone determination during milking by using a specific enzyme immunosorbent assay as a reference (chemical) method. Milk spectra with a wavelength range of 700 to 1050 nm and milk samples were collected every 20 s during milking from four lactating Holstein cows for 28 days using the NIR spectroscopic sensing system. Calibration models were developed using partial least squares analytical method and the precision and accuracy of the models was validated. Milk progesterone concentration for each milking was calculated by taking the progesterone concentration of the milk predicted values and milk yield obtained every 20 s, and was compared with the milk progesterone concentration chemical analysis value for one milking (bucket milk). The results obtained show that the measurement accuracy for one milking of milk progesterone concentrations was reasonably good. By installing the NIR spectroscopic sensing system developed in this study into an automatic milking system, it could predict milk progesterone concentration for one milking with almost the same accuracy as chemical analysis. Therefore, taking records of predicted values at every milking time and monitoring the continuous change of the milk progesterone concentrations, it would be possible to use this information for the detection of estrus status and diagnosis of pregnancy of each cow.

Cow Milk Progesterone Concentration Determination during Milking Using Near-infrared Spectroscopy

Patricia Iweka[1], Shuso Kawamura[*1], Tomohiro Mitani[2], Takashi Kawaguchi[3], Shigenobu Koseki[1]

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ABSTRACT

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Oral Session | Others (including the category of JSAM and SASJ)

[6-1015-D] Other Categories (3)

Chair:Takahiro Orikasa(Iwate University, Japan)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room D (4th room)

[6-1015-D-01] Field Representation and Path Planning for Robot Tractors

*Hao Wang[1], Noboru Noguchi[1] （1. Hokkaido University(Japan)）
10:15 AM - 10:30 AM

[6-1015-D-02] Driving Force Control for Suppression of Tractor's Dynamic Pitching Angle

*Yuya Aoyagi[1], Masami Matsui[2] （1. Tokyo University of Agriculture and Technology (Japan), 2. Utsunomiya University(Japan)）
10:30 AM - 10:45 AM

[6-1015-D-03] Development of a Smart Spraying System For Weeds On Rice Fields

*Thanh Tinh Nguyen[1], Ricardo Ospina[2], Noboru Noguchi[2] （1. Hokkaido University, Graduate School of Agriculture(Japan), 2. Hokkaido University, Research Faculty of Agriculture(Japan)）
10:45 AM - 11:00 AM

[6-1015-D-04] Deep Learning and Multiple Sensors Data Acquisition System for Real-time Decision Analysis in Agriculture Using Unmanned Aerial Vehicle

*Yunyan Xie[1], Ryozo Noguchi[2], Tofael Ahamed[2] （1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan)）
11:00 AM - 11:15 AM

[6-1015-D-05] Autonomous Navigation and Obstacle Avoidance for a Robotic Mower using Machine Vision

*Kosuke Inoue[1] （1. The University of Tokyo(Japan)）
11:15 AM - 11:30 AM

10:15 AM - 10:30 AM (Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room D)

[6-1015-D-01] Field Representation and Path Planning for Robot Tractors

*Hao Wang[1], Noboru Noguchi[1]　(1. Hokkaido University(Japan))
Keywords: Smart agriculture, Agricultural robots, Path planning, Headland turning, Minimum bounding box

An optimal coverage path planning method is presented to improve field efficiency; and in particular, to fully utilize the advantages provided by automatically guided farming equipment. In addition, several transfer paths are created to optimize the non-working distance and time consumption. To use the merit of agricultural robots, the backward movement along the navigation path is proposed in this research. Refiling or emptying the machine is not considered in this autonomous path planning. When a non-convex field is divided into several convex sub-fields, each sub-area has to be visited once without discard. The algorithm calculates an optimal working direction and order of sub-field to decrease the non-working area. In addition, the navigation path in this study consists of two parts; one is navigation points, which distribute in the area of the farm as an array. The other part is the code indicating the settings of the tractor and the operations at that position. The coding mechanism is informative enough for complex scenarios.

Oral Session | Others (including the category of JSAM and SASJ)
[6-1015-D] Other Categories (3)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room D (4th room)

[6-1015-D-02] Driving Force Control for Suppression of Tractor's Dynamic Pitching Angle

*Yuya Aoyagi[1], Masami Matsui[2] （1. Tokyo University of Agriculture and Technology (Japan), 2. Utsunomiya University(Japan)）

Keywords: Tractor, Traveling simulation, Torque control, Prevent accident, Suppression for attitude angle

In recent years, Japan's agricultural industry has had the highest mortality rate among all other industries. The agricultural industry has also become one of the most dangerous in other countries, and this is becoming a serious problem worldwide. In Japan, the highest number of fatalities (approximately 100 fatalities per annum) result from accidents involving tractors, with the most common being rollovers. Overturning accidents can occur either when a tractor is traveling along sloped, rough terrain because the uneven road surface increases the dynamic pitch angle beyond the overturning limit, or when the operator loses control as a result of the front wheels bouncing. To prevent these accidents, it is important to control the pitch angle when traveling along such terrain. In a previous study on the attitude stabilization of vehicles while they are being driven, attitude control technology using driving torque was developed. This technology improves riding comfort in automobiles traveling along paved roads. By applying such control to a tractor, it is expected that pitching overturn accidents can be prevented. In this scenario, it is important to confirm the dynamic effect on the pitching suppression by driving torque control. Overall, in this study, a three degrees-of-freedom, vertical, pitching, and forward/backward movement behavior model of a tractor that considers the influence of the driving force on the pitch angle was developed. The reaction force that each wheel received from the road surface was calculated, and numerical calculations were performed for the acceleration along each degree of freedom. The feedback control system had a static pitch angle on the input terrain as the target value, which was applied to the model. In the control system, using PID control, the driving force was calculated from each term (proportional, integrated, and derivative) of deviations, which consist of the difference between the target value and the dynamic pitch angle. The coefficients of the PID system were determined to become effective for this condition. The limits of the driving force were set according to the specification value of the tractor engine power and the power required for slope climbing. Driving simulation using the topographic information on a site of an actual tractor overturn accident was performed with both the driving-force control model and the constant-speed traveling model. The suppression of the pitch angle by controlling the driving force was examined through a comparative analysis of the results. The pitch angle of the driving-force control model was smaller than that of the constant-speed traveling model, enabling travel along the terrain. Thus, dynamic pitch angle control was implemented and validated. The maximum pitch angle of the driving-force control model was approximately 10% smaller than that of the constant-speed traveling model. In addition, a driving force corresponding to the attitude was generated within the usable driving-force range. Pitch angle suppression was confirmed by controlling the driving force. In our study, driving simulation using topographical information on the actual accident site was conducted to examine the suppression of the dynamic pitch angle by driving-force control. Consequently, the pitch angle of the driving-force control model enabled travel along the terrain. The maximum pitch angle of the control model was approximately 10% smaller than that of the constant-speed traveling model. The results suggest that the attitude angle can be suppressed by driving-force control to prevent tractor rollover accidents.

Driving Force Control for the Suppression of the Dynamic Pitching Angle of Tractors

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ABSTRACT

In recent years, Japan's agricultural industry has had the highest mortality rate among all other industries. Further, the agricultural industry has become one of the most dangerous in other countries; this is becoming a serious problem worldwide. In Japan, the highest number of fatalities (approximately 100 fatalities per annum) are due to accidents involving tractors, with the most common being rollovers. Overturning accidents can occur either when tractors are traveling along a sloped, rough terrain because uneven road surfaces increase the dynamic pitch angle beyond the overturning limit, or when the operator loses control owing to the bouncing of the front wheels. To prevent these accidents, the pitch angle should be controlled when traveling along such terrains. By applying attitude control using driving torque to a tractor, pitching overturn accidents could be prevented. In such scenarios, the dynamic effect on the pitching suppression by driving torque control should be verified. Overall, in this study, a three degrees-of-freedom, vertical, pitching, and forward/backward movement behavior model of a tractor that considers the influence of the driving force on the pitch angle was developed. The feedback control system had a static pitch angle on the input terrain as the target value, which was applied to the model. In the control system, using Proportional-Integral-Differential (PID) control, the driving force was calculated from each term (proportional, integrated, and derivative) of deviations, which comprises the difference between the target value and the dynamic pitch angle. The coefficients of the PID system were effective under this study's condition (mechanical specifications and topographical information, etc.). The limits of the driving force were set according to the specification value of the tractor engine power and power required for slope climbing. A driving simulation using the topographic information on a site of an actual tractor overturn accident was conducted with both the driving-force control and constant-speed traveling models. The suppression of the pitch angle by controlling the driving force was examined through a comparative analysis of the results. The pitch angle of the driving-force control model was smaller than that of the constant-speed traveling model, enabling travel along the terrain. Thus, dynamic pitch angle control was implemented and validated. The maximum pitch angle of the driving-force control model was approximately 10% smaller than that of the constant-speed traveling model. In addition, a driving force corresponding to the attitude was generated within the usable driving-force range. Pitch angle suppression was confirmed by controlling the driving force. In our study, a driving simulation using topographical information on the actual accident site was conducted to examine the suppression of the dynamic pitch angle by driving-force control. Consequently, the pitch angle of the driving-force control model enabled travel along the terrain. The maximum pitch angle of the control model was approximately 10% smaller than that of the constant-speed traveling model. The results suggest that the attitude angle can be suppressed by controlling the driving force to prevent tractor rollover accidents.

Keywords: Tractor, Traveling simulation, Torque control, Prevent accident, Suppression for attitude angle

1. INTRODUCTION

In recent years, with the development of agricultural machinery, working efficiency has considerably improved and labor burden significantly reduced. However, there are approximately 350 annual cases of fatal farm accidents in Japan, indicating a high accident rate (JMAFF, 2019). In addition, the fatal

injury rate per 100,000 farmers is as high as 24.0 people in the US (USDL, 2017), 16.1 people in Japan (JMAFF, 2015), and 9.2 people in the UK (HSE, 2019). Thus, in advanced countries with mechanized agriculture, agricultural injury rates are at a high level compared with other industries. This indicates that there may be more lives being lost in areas even beyond the scope of the surveys. The agricultural industry is becoming the most dangerous industry among all industries, increasingly becoming a serious problem worldwide. Therefore, taking adequate measures to reduce the fatalities is not only an important issue but also an urgent issue.

In Japan, the highest number of fatalities (approximately 100 fatalities per annum) is due to tractor accidents and the most common cause is overturning of tractors. Because of the terrain, most tractors in Japan have to travel on rough and steep slopes. This can increase the dynamic pitching angle with the risk of exceeding the overturn angle or steering control loss. To prevent such fall accidents of tractors, it is important to control the pitching angle when traveling on rough and steep terrains. This is achieved by applying the attitude control technology that inputs the driving torque to the tractor. However, it is important to verify theoretically the pitching suppression effect.

Previous studies on tractor behavior have analyzed the simulation of tractor stability on a rigid slope (Li et al., 2016) and impact dynamics model for nonlinear bouncing of tractors (Watanabe et al., 2017). Further, an analysis of tractor pitching based on an actual accident site (Matsui et al., 2016) has also been conducted. In addition, for vehicles traveling on paved roads, research on how to vary the drive torque and stabilize the vehicle attitude to improve driving comfort has been conducted (Takahashi et al., 2016, Sugai, 2016, Sawada et al., 2005). However, although there are studies on behavior stability analysis of tractors and posture stabilization in vehicles traveling on pavements, there are a few studies on posture stabilization in agricultural machines traveling on agricultural roads (rough and steep slopes). Therefore, in this study, a dynamic simulator of a tractor with three degrees of freedom (vertical, pitching, and horizontal) was developed, considering the influence of driving force on pitching. The feedback control system was applied to the model (dynamic simulator) and static pitching angle on the input terrain was set on the target value for the system. The driving force control and constant speed models were run using topographical information of the actual accident site, the results were compared, and the pitching angle suppression effect by the driving force control was examined.

2. MATERIALS AND METHODS

Dynamic equations with three degrees of freedom (vertical, pitching, and horizontal) for the tractor were formulated considering the influence of the driving force on the pitching. The behavior model of the tractor is shown in Fig. 1.
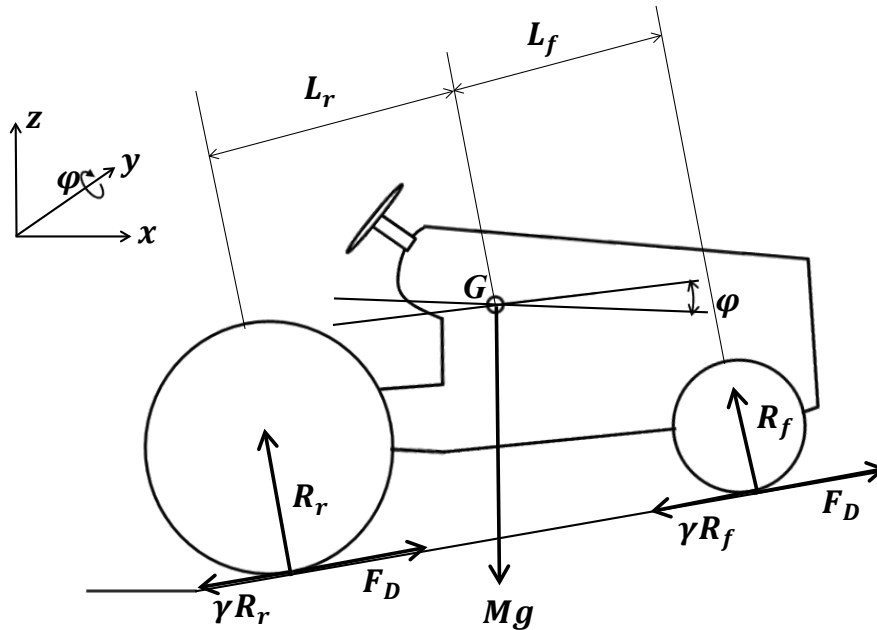

Figure 1. Behavior model of the tractor

Equations (1) to (3) provide the dynamic equations for the three degrees of freedom for the tractor. Equations (4) and (5) provide the wheel reaction and driving forces, respectively.

$$M\ddot{z} = Mg - (R_{fr} + R_{fl} + R_{rr} + R_{rl})\cos\varphi + \gamma(R_{fr} + R_{fl} + R_{rr} + R_{rl})\sin\varphi - F_D\sin\varphi \tag{1}$$

$$I_y\ddot{\varphi} = (R_{fr} + R_{fl})L_f - (R_{rr} + R_{rl})L_r - \gamma(R_{fr} + R_{fl} + R_{rr} + R_{rl})L_g + F_D L_g \tag{2}$$

$$M\ddot{x} = F_D\cos\varphi - \gamma(R_{fr} + R_{fl} + R_{rr} + R_{rl})\cos\varphi - (R_{fr} + R_{fl} + R_{rr} + R_{rl})\sin\varphi \tag{3}$$

$$R_{ij} = (z + L_i\varphi)\,k_i + (\dot{z} + L_i\dot{\varphi})\,c_i \tag{4}$$

$$F_D(t) = K_p e(t) + K_i \int e(t)dt + K_d \frac{de(t)}{dt} \tag{5}$$

where,

M = Mass of body [kg], F_D = Driving force [N], R = Reaction force of wheel [N],

I_y = Inertia moment of pitching [kg · m^2], k = Spring coefficient [N/m],

c = Damping coefficient [N · s/m], g = Gravity acceleration [m/s^2], z = Vertical displacement [m],

φ = Pitching angle [rad], x = Horizontal displacement [m],

L_f = Length of mass center to front axle [m], L_r = Length of mass center to rear axle [m],

L_g = Length of mass center to ground [m], $e(t)$ = Deviation of control system

Based on these dynamic equations, an original simulator using Matlab/Simulink was developed. In the simulator, the reaction force that each wheel receives from the road surface was calculated, and the acceleration for each degree of freedom was numerically calculated using the fourth-order Runge–Kutta method. The tractor specifications and model parameters are summarized in Table 1.

Table 1. Tractor specifications and model parameters

Item	Value
Body mass [kg]	1050
Full length [m]	2.98
Full width [m]	1.32
Total height [m]	1.93
Wheel base [m]	1.5
Distance of mass center and ground [m]	0.91
Distance of mass center and rear axle [m]	0.65
Kp [−]	100
Ki [−]	280
Kd [−]	20
Inertia moment of pitching [kg · m^2]	600
Front wheel spring coefficient [N/m]	60000
Front wheel damping coefficient [N · s/m]	200
Rear wheel spring coefficient [N/m]	160000
Rear wheel damping coefficient [N · s/m]	4400

The specifications used catalog values of a 30-horsepower tractor; further, the spring coefficient, damping coefficient, and moment of inertia used the values measured experimentally (Aoyagi et al., 2016).

A control system diagram is shown in Fig. 2. In the control system, using PID control, the driving force was calculated from each term (proportional, integrated, and derivative) of deviations, which comprises the difference between the target value and the dynamic pitch angle. As for control parameters, Kp, Ki, and Kd were searched in the range 1–500, respectively, and the most effective parameters of suppressing the maximum pitching angle were determined for the conditions assumed in this study. The limits of the driving force were set according to the specification value of the tractor engine power and power required for slope climbing.
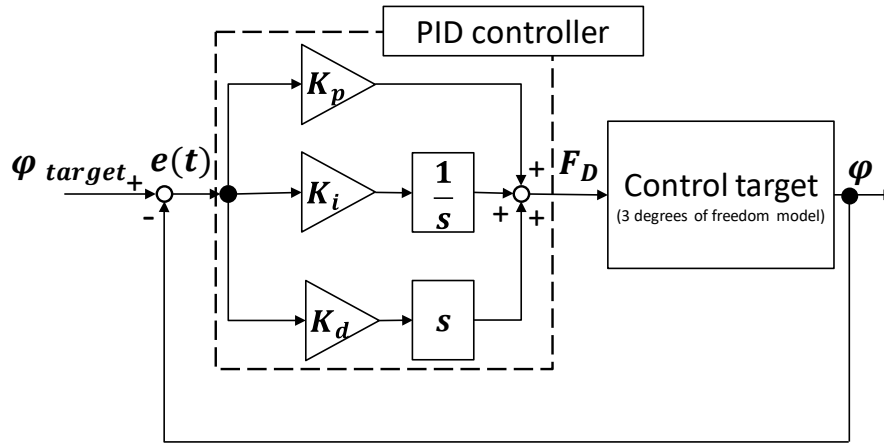
Figure 2. Control system diagram

For a simulation using the topographical information of actual accident site, we conducted a survey and hearing considering a tractor fall accident site that occurred in Niigata Prefecture in 2013 (Fig. 3).
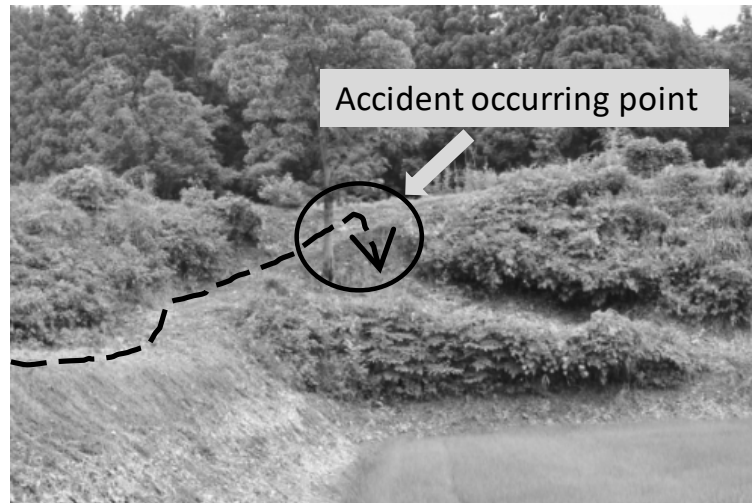

Figure 3. Accident site of the tractor

As for how the actual accident occurred, when the tractor climbed up on a farm road with a slope of about 20° at a traveling speed of about 1.0 m/s, the front wheel of the tractor bounced up and fell to the right near the middle of the slope. The input topography was reproduced by adding measured surface roughness to spline interpolation of the topography data obtained by surveying (Fig. 4).
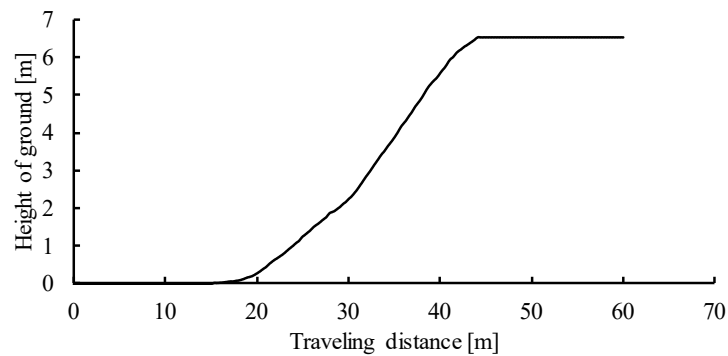
Figure 4. Input terrain

The driving simulation was conducted with the driving force control model and constant speed model, the results were compared, and the pitching angle suppression effect by the driving force control was examined.

3. RESULTS AND DISCUSSION

Fig. 5 shows the pitching displacement of the pitching angle control and constant speed models (1.0 m/s). The control effect (suppressed pitch angle) can be confirmed by observing that compared with the constant speed model, the pitching angle is suppressed for the control model. In particular, the control effect is remarkable in the traveling distance between 30 m and 40 m. The maximum pitching angle is 18.4° for the control model and 21.0° for the constant speed model around the traveling distance of 40 m. The maximum pitching angle of the control model is approximately 88% of the maximum pitching angle of the constant speed model.
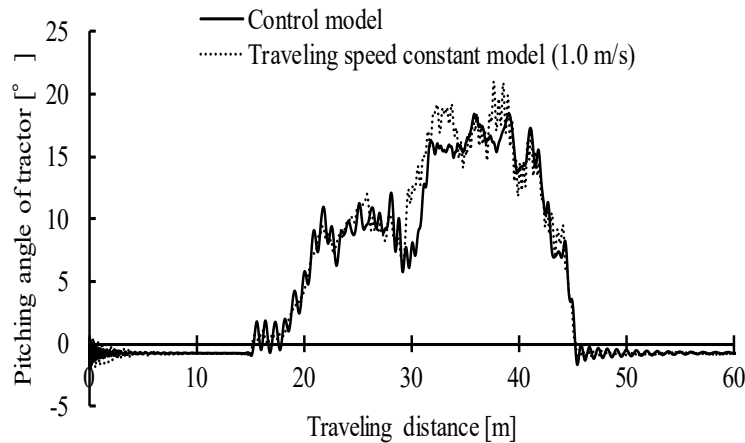
Figure 5. Pitching displacements of the control and constant speed models

Fig. 6 shows the increasing and decreasing of the pitching angle (the difference between the dynamic attitude angle and the static attitude angle) compared with the static pitching angle. Fig. 6 shows that the pitching angle is increased to a maximum of 3.7° in the control model and the pitching angle is increased at a maximum of 7.7° in the constant speed model. Moreover, the RMS value of each pitching angle increase/decrease in the travel distance between 15 m and 45 m (climbing section) is 1.9° in the control model and 2.6° in the constant speed traveling model; further, it travels more parallelly to the topography in the control model (showing a pitching angle close to the static attitude angle).
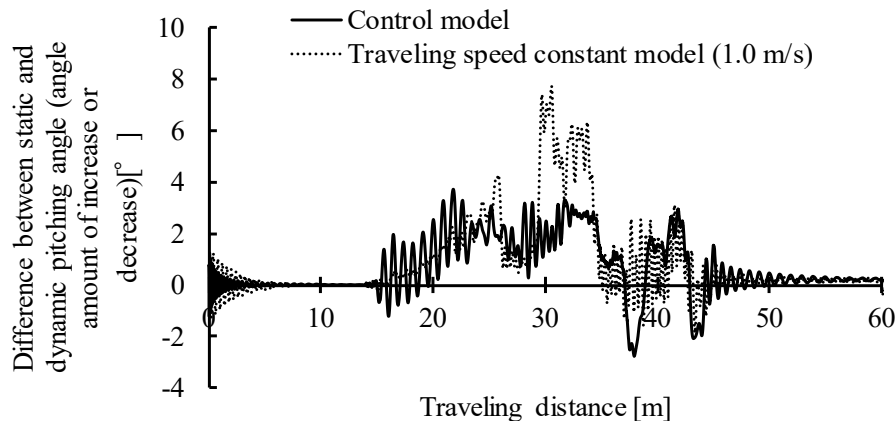
Figure 6. Fluctuations of pitching angle in comparison to static angle

Fig. 7 shows the driving force displacement of the pitching angle control model. The figure also shows that the driving force in the control model corresponds to the posture. This indicates the possibility of effectively controlling attitude by controlling the driving force according to the attitude angle.
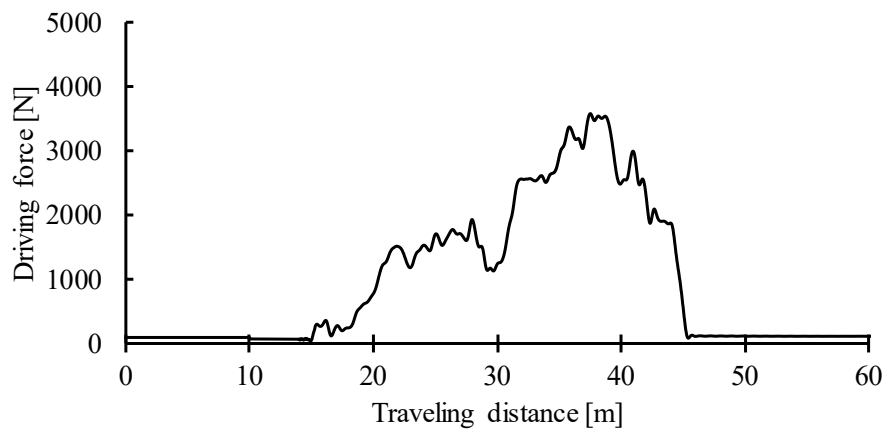
Figure 7. Driving force displacement of the control

From the above research, the suppression effect of the dynamic pitching angle by the driving force control is examined by the driving simulation based on topographical information of the actual accident site. The pitching angle suppression effect is obtained by controlling the driving force, and it was possible to travel along the terrain compared with the constant speed model. In addition, it was shown that the maximum pitching angle of the control model was approximately 10% lower than the maximum pitching angle of the constant speed model under the conditions of this study. From this, the possibility of attitude angle suppression by driving force control for fall accident prevention was shown.

4. CONCLUSION

Farm work accidents are a global issue. In Japan, the most common agricultural accident is the overturning of tractors, which require the most urgent mitigating measures. The stabilization of the posture of the tractor by drive torque control is expected to prevent pitching overturn, and it is important to theoretically verify the pitching suppression effect. In this study, a dynamic simulator with three degrees of freedom (vertical, pitching, and horizontal) of a tractor was developed considering the influence of the driving force on the pitching. The feedback control system was applied to the model (dynamic simulator) and static pitching angle on the input terrain was set on the target value for the system. The driving force control and constant speed models were run using the topographical information of the actual accident site; the results were compared, and the pitching angle suppression effect by the driving force control was examined.

As a result of this simulation, the pitching angle of the control model could be suppressed (traveling along the terrain) compared with that of the constant speed model, and the control effect can be confirmed. Furthermore, the maximum pitching angle is 18.4° for the control model and 21.0° for the constant speed model; the maximum pitching angle of the control model is about 10% lower than the constant speed model. In addition, the pitching angle of the control model rises 3.7° with respect to the static attitude and pitching angles of the constant-speed travel model rises at 7.7°, and the driving force according to the attitude is generated in the control model.

From the above research, the suppression effect of the dynamic pitching angle by the driving force control was examined by the driving simulation based on the topographical information of the actual accident site. The pitching angle suppression effect was obtained by controlling the driving force, and it was possible to travel along the terrain compared with the constant speed model. In addition, it was shown that the maximum pitching angle of the control model was about 10% lower than the maximum pitching angle of the constant speed model under the conditions of this study. From this, the possibility of attitude angle suppressing by driving force control for fall accident prevention was demonstrated.

ACKNOWLEDGMENT

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Oral Session | Others (including the category of JSAM and SASJ)
[6-1015-D] Other Categories (3)
Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room D (4th room)

[6-1015-D-03] Development of a Smart Spraying System For Weeds On Rice Fields

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Keywords: Precision agriculture, Weeds, Rice field, Image processing, Real time detection

In recent years, precision agriculture has become an important aspect of sustainable agriculture and environment protection. The problem of how to reduce the amount of chemicals used on the fields while ensuring productivity has become a challenge. In this study, we developed a smart spraying system prototype which uses a machine vision system capable of performing real time detection of two kinds of weeds in the paddy fields of the Vietnamese Mekong Delta (VMD). The prototype of the smart spraying system was tested to perform real time precision spraying of herbicide on weed location. The input images were recorded by using an RGB camera. The discrimination between rice and weeds was obtained by processing the images based on well-known image segmentation methods and analysis of the bounding rectangle of blob. This method is simple but effective, allowing to detect narrow leaf weeds and broadleaf weeds on the post-emergence stage of weeds with reasonable accuracy. Accuracy of spraying was also evaluated, spray volume and rate application were adapted and tuned for real work conditions on the field. Results suggest this system is more precise and reliable in comparison to current methods used in Vietnam.

Development of a Smart Spraying System For Weeds On Rice Fields

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ABSTRACT

In recent years, precision agriculture has become an important aspect of sustainable agriculture and environment protection. The problem of how to reduce the amount of chemicals used on the fields while ensuring productivity has become a challenge. In this study, we developed a smart spraying system prototype which uses a machine vision system capable of performing real time detection of two kinds of weeds in the paddy fields of the Vietnamese Mekong Delta (VMD). The prototype of the smart spraying system was tested to perform real time precision spraying of herbicide on weed location. The input images were recorded by using an RGB camera. The discrimination between rice and weeds was obtained by processing the images based on well-known image segmentation methods and analysis of the bounding rectangle of blob. This method is simple but effective, allowing to detect narrow leaf weeds and broadleaf weeds on the post-emergence stage of weeds with reasonable accuracy. Accuracy of spraying was also evaluated, spray volume and rate application were adapted and tuned for real work conditions on the field. Results suggest this system is more precise and reliable in comparison to current methods used in Vietnam.

Keywords: Precision agriculture, Weeds, Rice field, Image processing, Real time detection.

1. INTRODUCTION

Agriculture production in Vietnam is facing increasing environmental impacts due to large amounts of herbicide used in various species of crops. According to the International survey of herbicide resistant weeds (Heap, 2019), the types of herbicide and site application have been increasing year by year and continue to enlarge. This is because agriculture needs to increase productivity and quantity to cover the food demands for a population of more than 7 billion all over the world. Vietnam is an agriculture-based country and a top rice exporter in the world market. The country has 65 percent of cultivated land area ranging from the north to the south; especially in the Mekong Delta, which is the biggest cultivated region in Vietnam. The government has issued many especial policies oriented to increase rice quantity and productivity (Dung et al., 1999). However, using a large amount of pesticides causes damage to the environment, the human health and effects sustainable agriculture.

Some recent researches show hard conditions for detection of plants mixed with weeds by using deep learning CNN algorithms (Barrero et al., 2016; dos Santos Ferreira et al., 2017). However, these algorithms show low accuracy with small object detection and processing time is not good for real time applications. Thus, current research applications for crops only distinguish between weeds and crop rows. In this study, a machine vision method based on bounding blob that can classify crops and weeds is introduced. This method can detect two kinds of weeds native from the rice fields of the VMD. Besides, a real-time spraying prototype was developed for evaluation of the detection and spraying system.

2. MATERIALS AND METHODS
2.1. Image processing

The common weeds present in rice fields in Vietnam are not different from common weeds along Asia. There are several types of commercial herbicides available based on the biological characteristics of weeds. Typically, there are two main kinds of weeds present on the rice fields of the Vietnam Mekong Delta (VMD); which can be classified as broadleaf and narrow leaf (including grasses and sedges) types(Caton et al., 2010). There are three stages during the crop growth that allow application of the herbicide (IRRI, 2018). However, some stages require a high amount of herbicide; causing pollution of the environment. Sometimes weeds are mature, so it is necessary to apply a stronger type of herbicide at a higher rate. Higher concentrations of herbicide will affect and even may destroy the rice plants, particularly in the panicle initiation and flowering of rice stages. The field condition during the post-emergence stage of the crop growth is shown in Figure 1. At that time, the rice plant is in the stage of 7-10 days after sowing. Figure 1 shows not only the different colors, but also the different sizes between rice and weeds.

Figure 1. Weed post-emergence.

Figure 1 also shows how rice leaves and weed leaves do not overlap together. Therefore, for this particular research this is a good condition to apply image processing for segmentation between weed, soil and rice.

The image processing algorithm flow chart is shown on Fig. 2. OpenCV library for C++ language was used to make the program. Each step in the flow chart is explained in detail as follow.
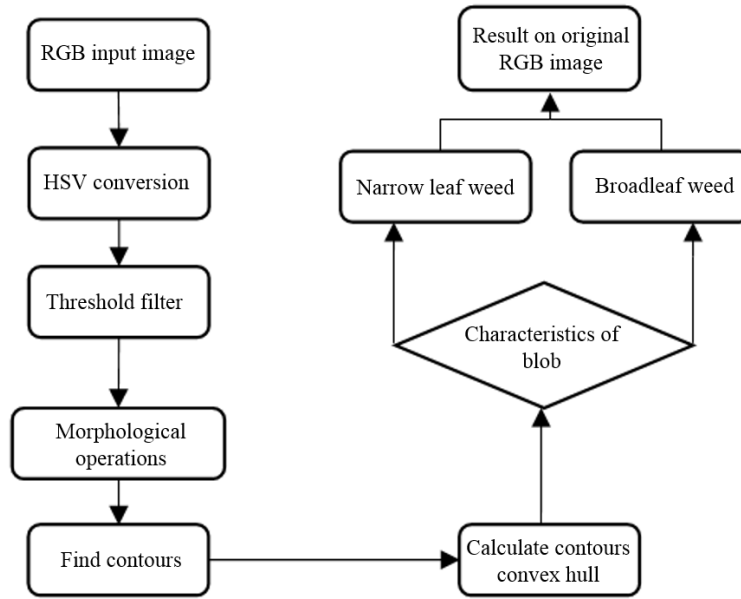
Figure 2. Algorithm of weed detection.

The input RGB image recorded by a camera is converted into the HSV color space for image segmentation. Then, a threshold filter is used to separate soil from vegetation, giving as a result a binary image. Morphological operations of dilation and erosion are applied to the binary image to smooth the noise and make the border of the plants look clear. Then, the contours of all the white regions on the image are detected. Calculation of the convex hull is performed on each detected contour in order to find the smallest convex set of points that can represent each contour on the image. Then, the bounding rectangle of each the contours is used to analyze the characteristics of each plant; as show in Figure 3. Such characteristics include the area of the bounding rectangle (S_r), the ratio between the height (h) and the width (w) of the bounding rectangle, the diagonal (d), and the ratio between the area of the contour (S_c) and its corresponding bounding rectangle. These characteristics help to remove the rice and remaining noises from the image. The setting values for detection between the narrow leaf and broadleaf weed are different. The ratio S_c/S_r is particularly helpful to distinguish the kind of weeds. Finally, the algorithm counts and labels each type of weed, with yellow for broadleaf weed and green for narrow leaf weed, as shown in Figure 4.
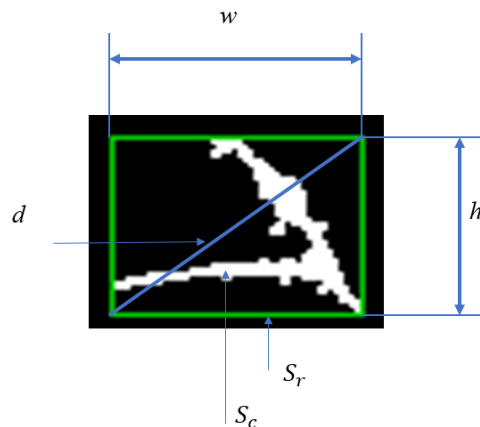

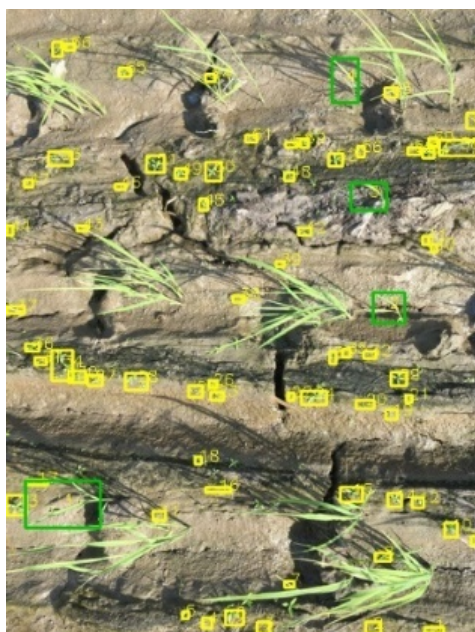Figure 3. Bounding rectangle characteristics for blob analysis.

Figure 4. Result of narrow leaf and broadleaf weed detection on original image.

2.2. Prototype of sprayer

A sprayer prototype was built for this study with the purpose of testing the effectiveness of the detection algorithm and explore the possibility of implementing into real life applications. Since the algorithm can detect two different types of weeds, two different types of herbicide can be applied. Figure 5 a) shows the full design in 3D and Figure 5 b) shows the real prototype. The prototype was built up on a steel table (0.9m length, 0.7m width, 0.8m height) which can move smoothly on 4 wheels placed on its legs.
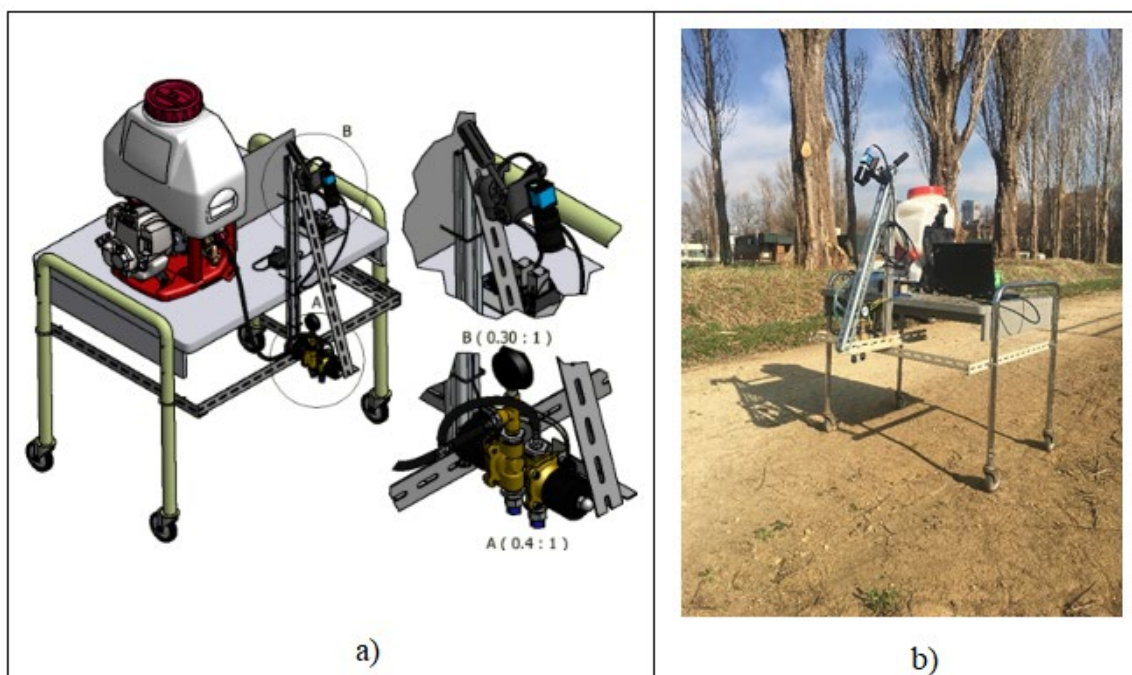
Figure 5. Sprayer prototype. a) Prototype on 3D design by Inventor software. b) Real prototype.

All the components and equipment of the prototype are shown on Figure 6; the camera is used to get image frames; an Arduino Uno is used as a controller and a backpack sprayer is used to control the spraying pressure. When the image processing algorithm detects the weed, the program will send a command to the Arduino Uno to activate the output signal to control the solenoid valve of the sprayer.
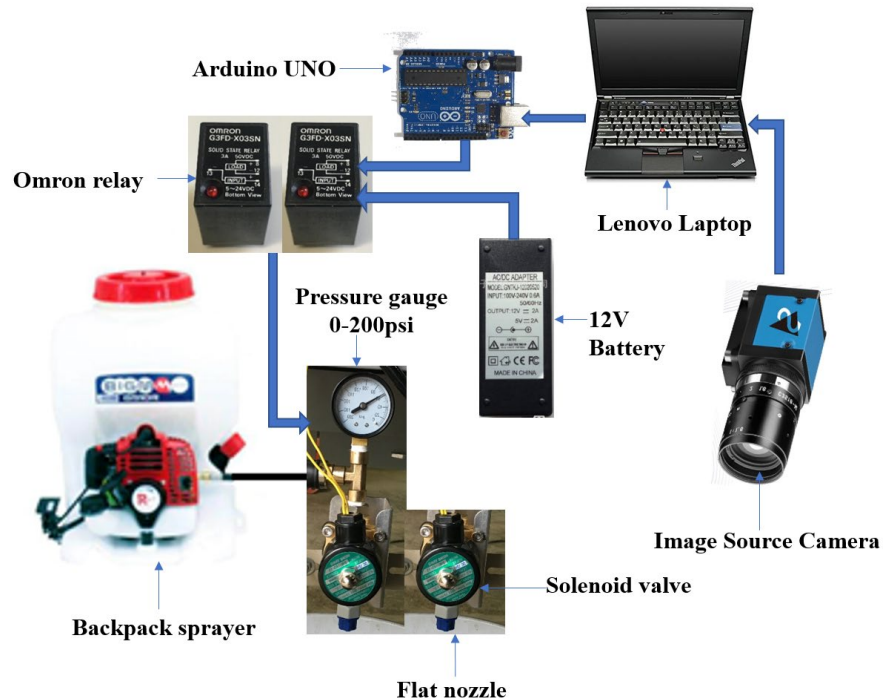

Figure 6. Accessories equipment and connection diagram of prototype.

3. RESULTS AND DISCUSSION

To evaluate the accuracy of the detection algorithm we used 112 RGB images, which were taken from real conditions on rice field in Vietnam. The real results were calculated for each sample picture based on an expert criterion, counting the weeds and rice plants. In this study, the linear regression method was used to evaluate the similarity between the detected values and the real values. Figure 8 a) shows the detection evaluation for broadleaf weed. Figure 8 b) shows the detection evaluation for narrow leaf weeds. In this experiment, it was show that the detection of broadleaf weeds has a higher accuracy than the detection of narrow leaf weed. The explanation for this result is because the broadleaf weed has a clear different size and shape compared to rice. However, the narrow leaf weed is not too much different.
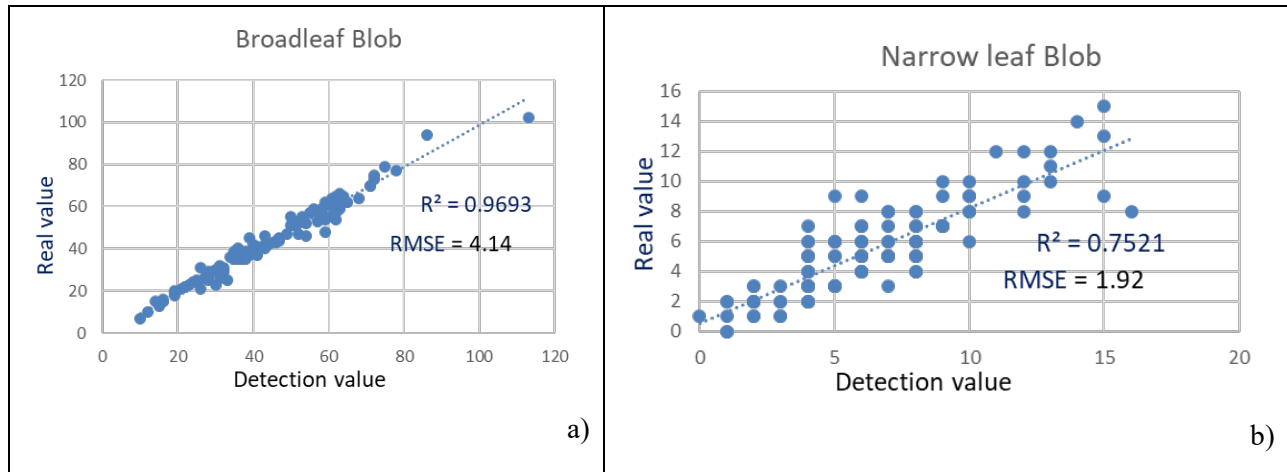
Figure 7. Weeds detection evaluation. a) Broadleaf weeds. b) Narrow leaf weeds.

For evaluation of the real time sprayer, the prototype was tested inside a laboratory because it is not suitable to use in the real rice field. The laboratory with cement floor can help to obtain a smooth movement without vibration, the indoor electric lighting system helps to balance light conditions for testing. Two types of weed samples were used. The size and shape of the samples is similar to the weeds found in Vietnam. The physical characteristics of the weed samples were carefully verified based on an expert criterion. The two kinds of the weed's samples were arranged in a random order in a straight line. Figure 9 shows the result of detection and spraying. When the prototype moves forward at 0.67 m/s, the nozzles spray at the correct position on each weed location; the water amount of each sprayer is 0.0048 liter. The resulting frame rate for the image processing algorithm is 10 FPS (Frame per second), and the delay time from the detection to nozzle sprayer activation is 0.35 seconds. Considering the traveling speed of the prototype, this method is suitable for effective spraying in real life applications. Besides, it is also consistent with the speed of rice transplanter machines traveling in paddy fields (Sato et al., 1996).

Figure 8. Experiment result of two kinds of weeds detection and spraying.

4. CONCLUSION

The proposed combined strategy works properly when the weeds are present on an early stage of growth, which coincides with the right timing for herbicide application. If the crop is further developed, the weed

will most likely present overlapping with the leaves of rice plants and the segmentation process will become difficult. Nevertheless, the proposed approach provides a useful methodology to discriminate weeds in the real paddy field situations, which display a mixture of rice, soil and weeds over many kinds of fields used in Vietnam like transplanting field, spot seeding field, and broadcasting field. Although the results can be considered satisfactory, this method might be only applied for distinguish weeds and rice on spot seeding fields and transplanting fields. In the broadcasting fields with high density of rice, the segmentation will be low, and results might not be accurate. It is also necessary to design strategies to cope with the variability of lighting conditions, and velocity and vibration of the spraying system traveling forwards. As future work, it is proposed to improve the image processing time. Also, a new mechanical design is required to improve the balance and reduce the vibration on the camera.

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11:00 AM - 11:15 AM (Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room D)

[6-1015-D-04] Deep Learning and Multiple Sensors Data Acquisition System for Real-time Decision Analysis in Agriculture Using Unmanned Aerial Vehicle

*Yunyan Xie[1], Ryozo Noguchi[2], Tofael Ahamed[2]　(1. Graduate School of Life and Environmental Sciences, University of Tsukuba(Japan), 2. Faculty of Life and Environmental Sciences, University of Tsukuba(Japan))
Keywords: UAV, Machine Learning, Deep Learning, Multiple Sensors

This research was conducted to develop a user-friendly application to connect multiple sensors while using UAV to collect field data. The onboard and ground sensors were connected in the same application for ease of data collection in one software application. In the onboard sensors, thermal and RGB cameras were connected and transmitted the images within 500 m – 1000 m range. The soil moisture content information, humidity information were collected. In addition, the image analysis and deep-learning algorithm was added to the classification of the objects while landing. Histogram of Oriented Gradient (HOG) and Support Vector Machine (SVM) and YOLOV3 algorithms were implemented for classification of human, vehicle and others obstacle. The Michihibiki module was also connected with IoT application to soil moisture content measurement in the larger fields. The user application is divided into three modules: Hardware Module for Sensors Networks (HMSN), Software Module for Data Acquisition (SMDA), and Deeping Learning for Decision Analysis (DLDA). This research will be extended further with real-time analysis and decision support systems for UAV-based agricultural operations and safety systems.

Oral Session | Others (including the category of JSAM and SASJ)

[6-1015-D] Other Categories (3)

Fri. Sep 6, 2019 10:15 AM - 11:30 AM Room D (4th room)

[6-1015-D-05] Autonomous Navigation and Obstacle Avoidance for a Robotic Mower using Machine Vision

*Kosuke Inoue[1]　（1. The University of Tokyo(Japan)）

Keywords: Autonomous Navigation, Visual SLAM, Obstacle Avoidance, Deep Learning, Object Detection, Stereo Camera

The autonomous operation of agricultural machinery using global navigation satellite system (GNSS) information has recently experienced rapid development as a labor-saving measure in agriculture. The self position is recognized with a GNNS signal, and the vehicle can travel in the area autonomously. However, if the vehicle is driven using only the GNSS signal such that the surrounding environment is not recognized, there is a risk of collision with an obstacle. Furthermore, sensors such as radars or lasers cannot distinguish between grass and obstacles and thus cannot be used to detect the likely obstacles encountered by agricultural machinery. Autonomous driving cannot be performed in environments where the satellite positioning accuracy is low, such as orchards. Herein, an autonomous driving system was developed that performs obstacle avoidance and autonomous driving without a GNSS signal by using an object detection system that is based on a stereo camera and deep learning. Stereo cameras and convolutional neural networks recognize the environment and avoid obstacles. The self position is corrected by observing a landmark in the environment. The experiment will be conducted at the Tanashi Forest of the University of Tokyo to evaluate autonomous driving by employing real-time kinematic-GNSS to measure the true values.

Autonomous navigation and obstacle avoidance for a robotic mower using machine vision

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ABSTRACT

The autonomous operation of agricultural machinery using global navigation satellite system (GNSS) information has recently experienced rapid development as a labor-saving measure in agriculture. The self position is recognized with a GNNS signal, and the vehicle can travel in the area autonomously. However, if the vehicle is driven using only the GNSS signal such that the surrounding environment is not recognized, there is a risk of collision with an obstacle. Furthermore, sensors such as radars or lasers cannot distinguish between grass and obstacles and thus cannot be used to detect the likely obstacles encountered by agricultural machinery. Autonomous driving cannot be performed in environments where the satellite positioning accuracy is low, such as orchards. Herein, an autonomous driving system was developed that performs obstacle avoidance and autonomous driving without a GNSS signal by using an object detection system that is based on a stereo camera and deep learning. Stereo cameras and convolutional neural networks recognize the environment and avoid obstacles. The self position is corrected by observing a landmark in the environment. The experiment will be conducted at the Tanashi Experimental Forest of the University of Tokyo to evaluate autonomous driving by employing real-time kinematic-GNSS to measure the true values.

Keywords: Autonomous Navigation, Visual SLAM, Obstacle Avoidance, Deep Learning, Object Detection

1. INTRODUCTION

The automation of farm operations using robot technology has increased in Japan due to the decreasing number and aging of farmers. Autonomous tractors, which can automatically perform agricultural tasks using GNSS information, have been developed. We are currently developing a robot mower that uses GNSS information. While it is possible to estimate the self position using the high-precision positioning from the GNSS signal and automatically conduct agricultural tasks in the designated area of a field, there is a risk of collision with an obstacle since the surrounding area cannot be recognized by automated driving that is solely based on the GNSS signal. Obstacles are recognized using sensors such as lasers and radar for the autonomous travel of automobiles and robots. However, these sensors cannot identify the type of object, and the resulting robot mower cannot distinguish between grass and other obstacles during autonomous operations. Furthermore, high-precision GNSS positioning cannot be used in orchards and other places where satellite signals are blocked, rendering GNSS-based autonomous driving inoperable.

Herein, we develop a system for detecting obstacles and estimating the position of objects using image recognition. Obstacles in the environment are recognized using object detection with a convolutional neural network (CNN), the self position is estimated using Visual SLAM, and the obstacle avoidance is performed by combining the self-position and object detection information. A landmark with a known position is installed in the environment to improve the accuracy of the self-position estimation, with the self position corrected based on the landmark location.

2. MATERIALS AND METHODS

Herein, we combined the self-position estimation from Visual SLAM, object detection via deep learning, and obstacle avoidance from the path optimization Self-Position Estimation.

2.1 Hardware

2.1.1 Robot configuration

The robot that has been designed for this study is shown in Figure 1. The rear wheels are controlled by sending a signal from the motor driver. An embedded system is contained in the box, which is used for

environment recognition and route generation, with the driving commands sent from the embedded system to the motor driver.

Two cameras are used as the sensors and are mounted in the front and rear of the autonomous driving system on the assumption of round cutting. An inertial measurement unit is also mounted to the system to improve the accuracy of the self-position estimation.

Figure 1. Robot used in this study.

2.1.2 Camera

Intel Realsense D435 is used for depth camera (Figure 2). Realsense D435 comes with an RGB camera and two near-infrared cameras that capture depth images via stereo matching.

Figure 2. Realsense D435 depth camera.

2.1.3 Embedded system

Nvidia Jetson Xavier is used for the embedded system. Jetson Xavier is equipped with a graphics processing unit (GPU) and can be used for deep-learning applications.

2.2 Software

2.2.1 Self-position estimation

We use real-time appearance-based mapping (RTAB-map), which is a Visual SLAM library, to estimate the self position. The feature points are extracted from consecutive images in Visual SLAM, and the amount of movement between frames is estimated by matching the next image with feature points.

2.2.2 Obstacle and landmark detection

Yolo v3, which is an object detection algorithm with a CNN, is used for obstacle and landmark detection. Yolo v3 divides an image into grids and estimates the type of object and size of the bounding box for each grid. Here images of trees were collected from the Internet, and annotation data were manually created. Figure 3 illustrates this tree detection example using Yolo v3.
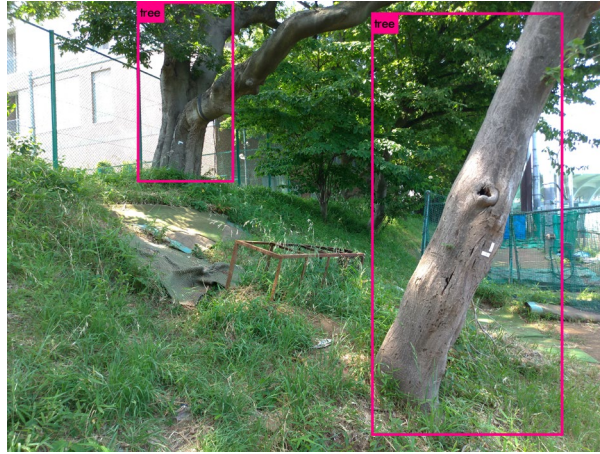
Figure 3. Tree detection example.

2.2.3 Position estimation of the object

Yolo v3 is used in conjunction with a CNN for the obstacle and landmark detection. The positions of the obstacles and landmarks are detected by the CNN, and the position of the detected object relative to the robot is estimated by associating the depth image with the detected position in the image. When an object is detected via the CNN, the distance is estimated by taking the center value of the distance, which corresponds to the pixel included in the detection position of the depth image, and the direction is estimated as the coordinate of the pixel in the center of the detection position and the camera parameter.

2.2.4 Obstacle avoidance path

Teb Local Planner is used for the avoidance route generation. The obstacle avoidance path is generated by rewarding the scheduled path and penalizing the obstacle to optimize the travel path. Examples of obstacle detection and an avoidance path are shown in Figure 4.
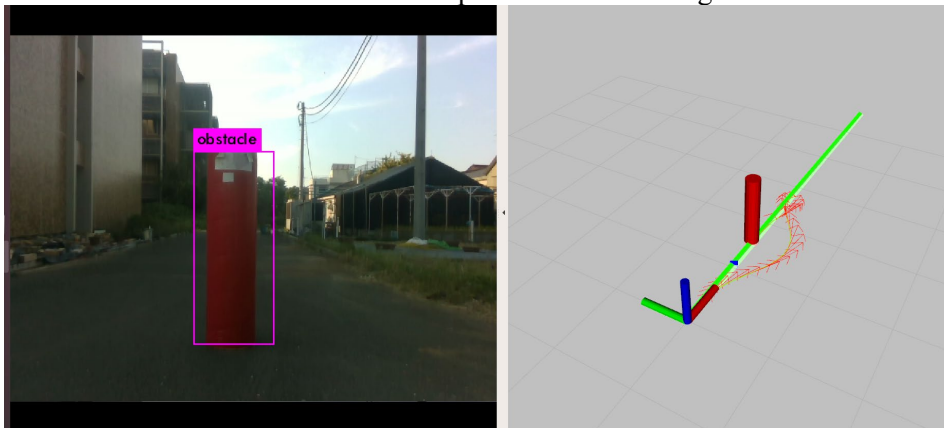

Figure 4. Examples of Obstacle Detection (left) and Avoidance (right).

2.2.5 Correcting the estimated position with a landmark observation

The drift of the estimated position is reduced by using the landmark in the CNN. The robot's position is estimated by observing a landmark with a known position. The self position is then corrected by adding the information on the estimated position to the appropriate node in the SLAM and adjusting the frame of reference accordingly.

3. EXPERIMENT

The experiment will be conducted at the Tanashi Experimental Forest, Graduate School of Agricultural and Life Sciences, the University of Tokyo. The accuracy evaluation of the self-position estimation will be performed by placing obstacles and landmarks in the test location, running the autonomous system in manual and autonomous mode, and measuring the true value using real-time kinematic (RTK)-GNSS.

4. ACKNOWLEDGMENT

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