Poster Session | Crop Genetics and Physiology | P4: Poster Session

[P4] Crop Genetics and Physiology

Thu. Sep 9, 2021 12:15 PM - 2:00 PM Room 4 (Poster) (Crop Genetics and Physiology)

1:15 PM - 2:00 PM

[P4-26]The Purification of Recombinant TGW6, which Limits Grain Size in Rice

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THOUSAND-GRAIN WEIGHT 6 (TGW6) encodes an indole-3-acetic acid (IAA)-glucose hydrolase. By the function of native TGW6, the number of endosperm cells and the weight of grains are limited. Otherwise, the 1-bp deletion allele of tgw6 cloned from the Indian landrace rice Kasalath loses the function and enhances the grain size as well as yield. We hypothesized that the chemical intervention strategy for the specific inhibition of TGW6 might increase the grain size and yield. However, we do not have the information for the structure of the TGW6 protein to design the antagonist. In this study, we purified the recombinant TGW6 through the Escherichia coli expression system. We cloned full-length TGW6 from Nipponbare by PCR and inserted it into pET-32b. The constructs were transformed into Rosetta-gami 2 (DE3). The E.coli cells contained a pET-32b expression plasmid for TGW6 were grown at 37°C in LB minimal medium. Isopropyl-1-thio-β-d-galactopyranoside was added to induce the expression of the TGW6 construct. After harvesting the cells, we extracted the recombinant TGW6 by sonication and purified using Ni-affinity chromatography. However, most of the recombinant TGW6 expressed as insoluble forms. Then, we cloned TGW6 truncated 30 amino acid sequences from Nterminus with the same experimental condition. We could improve the solubility by truncation of Nterminus amino acids and purify the recombinant TGW6. Further, we will report on the enzyme activity of the recombinant TGW6.