Digital holographic microscopy for quantitative observation of birefringent material in biological samples

Dept. of Applied Physics, Graduate School of Engineering, Osaka Univ.¹, Biophotonics Lab., Immunology Frontier Research Center, Osaka Univ.², [°]Wataru Kamo^{1,2}, Nicolas Pavillon², Katsumasa Fujita¹, and Nicholas Smith² E-mail: nsmith@ap.eng.osaka-u.ac.jp

Digital Holographic Microscopy (DHM) is a microscopic imaging technique based on interferometry. Since DHM digitally records a hologram generated by interference between object and reference beam, both amplitude and quantitative phase images can be obtained by numerically reconstructing the information of the wavefront of the light transmitted through the sample. DHM has been widely used for quantitative observation of live cell in a non-invasive way, and enables the extraction of parameters such as morphologies or dry-mass estimation, typically with picogram sensitivities by interpreting their phase as thickness [1]. As in classical wide-field microscopy, it is possible to make the measurement more specific by introducing a polarization selection in the detection scheme, such as to detect specific birefringent targets [2].

Here we developed a technique to obtain quantitative phase images from birefringent materials as well by combining the DHM system with a four independent cross-polarized measurement scheme. Classically, polarization light microscopy has been used for obtaining images of birefringent materials, but quantitative assessments are usually difficult. In the case of DHM, it is possible to retrieve quantitative information from birefringent materials, by measuring the phase delays induced by the optically anisotropic crystals, which are orientation-dependent. We can include these measurements within a Jones matrix formalism, which provides the full birefringent complex information of the measured material. In this presentation, we will discuss about implementation of our DHM system. Furthermore, we will demonstrate imaging results of living macrophage cells and birefringent malaria byproduct as an application, where polarization can then provide target-specific images.

- [1] B. Rappaz, E. Cano, T. Colomb, J. Kühn, C. Depeursinge, V. Simanis, P. J. Magistretti, and P. Marquet, "Noninvasive characterization of the fission yeast cell cycle by monitoring dry mass with digital holographic microscopy.," *J. Biomed. Opt.*, vol. 14, no. 3, p. 034049 (2009)
- [2] Y. Kim, J. Jeong, J. Jang, M. W. Kim, and Y. Park, "Polarization holographic microscopy for extracting spatio-temporally resolved Jones matrix," *Opt. Express*, vol. 20, no. 9, p. 9948 (2012)