Revealing Molecular Structure and Orientation with Stokes Vector Resolved Second Harmonic Generation Microscopy

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Abstract: We report on measurements and characterizations of polarization properties of Second Harmonic (SH) using a four-channel photon counting based Stokes polarimeter. In this way, the critical polarization parameters can be obtained concurrently without repeating imaging scanning at different acquisition time. The critical polarization parameters, including the degree of polarization (DOP), the degree of linear polarization (DOLP), and the degree of circular polarization (DOCP), are extracted from the reconstructed Stokes vector based SH images in a pixel-by-pixel manner. The measurements are further extended by varying the polarization states of the incident light and detecting the resulting Stokes parameters of the SH signal, to reveal the molecular structure and orientation of the samples. SH is known to sensitize the anisotropy of the index refractive tensor. The use of Stokes parameters is indispensable in identifying the proportion of fully polarized light and the corresponding phase shift between the eigen-polarization vectors. By applying the Stokes parameters based image analysis techniques SHG images of biomolecules, such as potato starch, collagen, and skeletal muscle. The combination of SH and Stokes polarimeter makes a powerful tool to investigate the structural order of targeted specimens. The observation from the polarization parameters reveals that these biomolecules are highly anisotropic, coincides with the known pitches of distinct helices within the coil structures of fibers. We have demonstrated that anisotropy and chirality from biological samples is accounted for in polarization resolved SHG imaging through DOLP and DOCP.

KEY WORDS: Stokes vector, second harmonic generation, Non-linear optical microscopy.

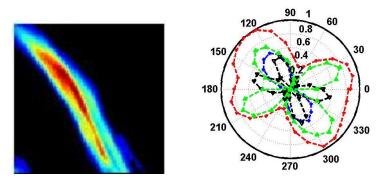


Fig. shows the SHG image of collagen fiber and Shows the graphs for absolute values of Stokes parameters as a function of incident polarization

References-

^{1.} P. J. Campagnola and L. M. Loew, "Second harmonic imaging microscopy for visualizing biomolecular arrays in cells, tissues and organisms," Nat. Biotechnol. 21, 1356-1360 (2003).

^{2.} L. M. Sandvik Aas, P. G. Ellingsen, and M. Kildemo, "Near infra-red Mueller matrix imaging system and application to retardance imaging of strain," Thin Solid Films 519, 2737–2741 (2011).

^{3.} M.R.Foreman, C.M.Romero, and P. Torok, "A priori information and optimization in polarimetry," Opt. Express 16, 15212-15227 (2008).
4. N. Mazumder, J. Qiu, M. R. Foreman, C. M. Romero, C.W. Hu, H. R. Tsai, P. Török, and F. J. Kao, "Polarization-resolved second harmonic generation microscopy with a four-channel Stokes-polarimeter," Opt. Express, 20, 14090-14099 (2012).