Fast Wide-field Polarimetric Second-Harmonic Generation Microscopy

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Polarimetric Second-Harmonic Generation (P-SHG) microscopy enables non-invasive imaging of non-centrosymmetric biological tissues. P-SHG provides detailed ultrastructural information on composition and organization of collagenous and muscular samples, beyond the diffraction limit. The extracted information is often in the form of second order nonlinear susceptibility ratios. This work pertains to the development of a wide-field P-SHG technique, which greatly enhances imaging speed, by using circularly polarized excitation and emission fields.

The imaging was performed using a high-power amplified laser (Pharos, Light Conversion), coupled to a custom wide-field nonlinear microscope, which enables fast imaging of large fields of view ($650\mu m \times 650\mu m$). A unique combination of linear and circular polarization states of the fundamental beam, and the corresponding linear and circular polarization measurements of the SHG signal allows for the extraction of second order susceptibility ratios, such as $R = \chi^{(2)}_{zzz}/\chi^{(2)}_{zxx}$ (xz-plane is the image plane), as well as other quantities such as SHG circular and linear dichroism (SHG-LD and SHG-CD) [1]. The analysis of the acquired images is completed in a few minutes, resulting in pixel-resolved parameter images, as shown in Fig. 1, that are invaluable in quantitative analysis of biomaterials.

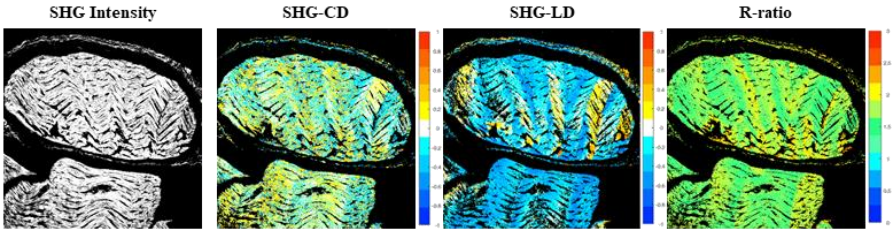

Figure 1: Polarimetric Second Harmonic Generation Microscopy of Pig Tendon

Wide-field P-SHG technique enables fast and precise investigations of ultrastructural variations in the biological tissues, with applications reaching beyond general tissue characterization. Fast imaging of large sample areas is extremely useful in biomedical diagnostics, contractility research, and histopathology [2,3]. In addition, the system was utilized in whole slide imaging of histopathology samples, aiding in high-throughput cancer diagnostics.

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