

Thickness dependences of anisotropic tissues on polarized second harmonic imaging

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ABSTRACT

Second-harmonic generation (SHG) microscopy is efficient in investigating the three-dimensional (3D) organization of anisotropic biological tissues, such as dermis, tendon and blood vessels. Among these, tendon is mainly composed of type-I collagen, which is abundant and structurally birefringent in vertebrates and makes the incident light splitting into two-cross polarized electric fields (i.e., ordinary and extraordinary wave). When performing SHG tissue imaging, linear polarized excitation is usually exploited to derive full structural information of the sample. However, special attention should be paid on the depolarization effect occurring in thick tissues because the phase retardation between the two field components changes with light penetration depth and refractive index difference due to birefringence. Consequently, a well-defined polarization state is slightly changed through the scattering process and the attainable penetration depth is restricted [1].

It has been reported that the degree of polarization of circularly polarized light maintains its initial polarization state for longer penetration depth than that of linearly and elliptically polarized light, which has been simulated with a polarization-sensitive Monte Carlo model [2]. In this work, to study how polarization states are influenced by sample thickness and compare the degree of light attenuation among different polarized excitations, we use linear, elliptical and circular polarized light to image multiple tendon samples of varying thickness. The difference of SHG signal affected by anisotropic tissues as well as penetration depth caused by the three types of polarizations will be verified and compared with the existing results.

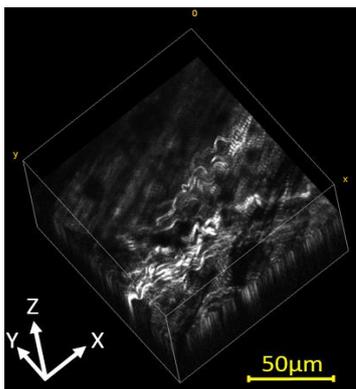


Figure 1: Volumetric SHG image of pork's tendon under the excitation of circular polarization. Size of the image stack: $138 \times 138 \times 42 \mu\text{m}^3$.

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