

## A NICHE FOR ULTRAFAST LASERS IN PROTEIN STRUCTURE DETERMINATION

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**Abstract.** The three-dimensional structures of large proteins and protein complexes are most commonly determined by X-ray diffraction of protein crystals. Consequently, protein crystallization is an essential and ubiquitous method in structural biology. A key practical complication associated with crystal structure determination stems from the vast phase-space of crystallization conditions possible coupled with the finite quantity of purified protein available. Currently, a crystal must be  $>10\ \mu\text{m}$  across to be definitively detected using methods currently in place for crystallization screening (e.g., based on birefringence, uv fluorescence, or image analysis), which corresponds to a volume of  $\sim 1000\ \mu\text{m}^3$ . Second harmonic generation microscopy using an ultrafast laser was found to be remarkably selective for detection of chiral crystals, including protein crystals, allowing detection of single crystals at sizes orders of magnitude lower than methods currently in place for crystal screening. Based on preliminary measurements, SHG can be expected to yield detectable signal to noise for crystals as small as  $\sim 100\ \text{nm}$  across, yielding a million-fold reduction in the minimum required detectable crystal volume, and correspondingly the total amount of protein needed to assay a particular set of conditions. Furthermore, second order nonlinear optical imaging of chiral crystals (SONICC) requires no special sample preparation and is directly compatible with most common crystallization platforms. Preliminary measurements and applications of this approach will be described.

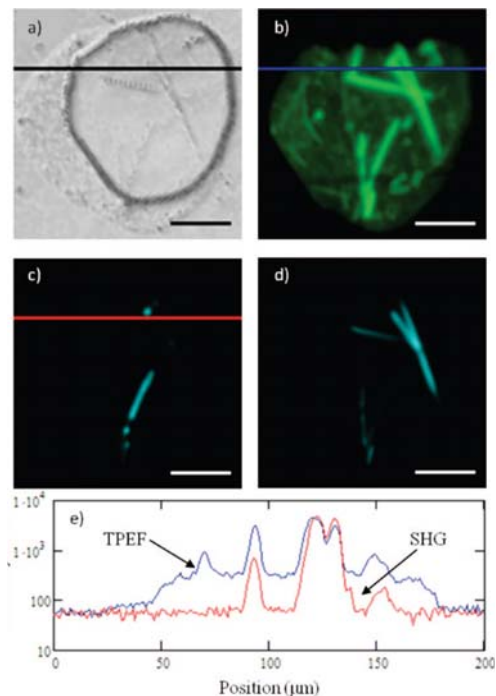


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