THE NONLINEAR GUIDE-STAR CONCEPT: A FAST AND NONINVASIVE METHOD TO MEASURE AND CORRECT SAMPLE INDUCED ABERRATIONS

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1. ABSTRACT

Adaptive optics (AO) was developed to restore the performance of optical systems (i.e. astronomical telescopes) affected by distortions produced to light as it travels through inhomogeneous media. This is carried out by measuring the distorted wavefront (WF) and then compensating for it, using an adaptive element. As any other imaging device, two-photon excited fluorescence (TPEF) microscopy is similarly affected when excitation light enters into a biological specimen which constraints its diffraction limited performance. Such effects have been successfully described by theoretical models using simple interfaces [1]. However, these cannot be applied to biological samples given its structural complexity. The implementation of a direct WF sensing scheme is not an easy task [2], therefore aberrations have been compensated using sensor-less approaches. These are based on the optimization of a merit function, where an adaptive element is shaped iteratively, thus requiring multiple images, being prone of causing photo-damage effects. We propose the use of the nonlinear guide-star (NL-GS) concept, a practical and fast technique to compensate for WF distortion. This methodology does not require any additional sample preparation (i.e. introduction of fluorescent beads) as it is based on the ability of TPEF to generate a confined focal volume at any position inside the sample. This incoherent emission is then used to capture the WF distortion produced by the sample inhomogeneous structure using a WF sensor [3]. Such information is then used to shape a deformable mirror in a single step. The NL-GS concept is demonstrated for *in vivo* biological samples showing its potential to improve the total collected signal intensity up to one order of magnitude.

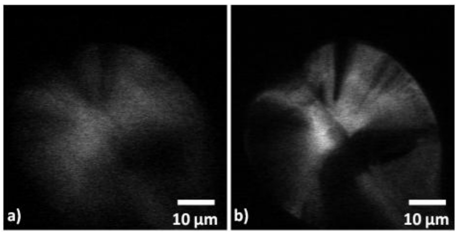

Figure 1: Single frames taken inside an *in vivo C. elegans* pharynx using TPEF microscopy. a) aberrated image. b) aberration compensated image.

2. REFERENCES

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