

Wide-field adaptive optics without guide stars

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1. INTRODUCTION

Sample-induced aberrations generally lead to reduced image quality in optical microscopy. A standard approach to counter such aberrations is to use adaptive optics (AO), which was first developed in astronomy but is now gaining traction in microscopy. The basic idea of AO is to insert an active optical correction element, typically a deformable mirror, in the optical path of the microscope to compensate for the aberrations produced by sample. The most common placement of this correction element, by far, is in a pupil plane of the microscope optics, called pupil AO. However, as first recognized by the astronomy community, a placement of the correction element in a plane conjugate to a primary sample aberration plane can lead to a significant field of view (FOV) advantage when these aberrations are spatially varying [1]. More recently this advantage of conjugate AO has been recognized by the microscopy community both in simulation studies and in experiment. In this paper we describe a novel implementation of conjugate AO, bearing in mind that our results can be equally applied to pupil AO.

2. TECHNIQUE

We demonstrate an implementation of widefield microscopy with sensor-based AO that does not require the use of guide stars [2]. Wavefront sensing is performed using illumination provided directly by the object itself, over the entire FOV of the wavefront correction. Since our implementation here involves conjugate AO (as opposed to pupil AO), the correction FOV is almost as large as the full FOV of our microscope. The development of our technique addressed two key challenges. The first challenge was the development of a wavefront sensor that exhibits large dynamic range capable of operating with relatively uncollimated light. For this we used a technique called partitioned aperture wavefront (PAW) sensing, previously developed in our lab for quantitative wavefront sensing [3]. The second challenge was to modify PAW sensing to enable it to work with an arbitrarily distributed extended source, namely the object itself. As we will see, this required supplementing PAW sensing with additional information provided by the science camera in our system (i.e. the imaging camera focused on the object), and applying a phase retrieval algorithm based on the Van Cittert-Zernike theorem.

We provide proof of principle demonstrations of our technique

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Wavefront Control by GPC

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1. GENERALIZED PHASE CONTRAST (GPC)

Sculpting the wavefronts of light in both fixed and programmable shapes has a variety of applications in both research, industry and medicine. With the widespread use of lasers that lend themselves to efficient reshaping due to their high spatial coherence, the versatility of wavefront control is further increased. Therefore, laser beam shaping based on photon-efficient phase-only methods are extensively applied in research such as in advanced adaptive and active microscopy and contemporary optical micro-manipulation [1,2] to mention a few typical uses. Phase-only light shaping and wavefront control is also finding its use in new and exciting applications such as for emerging neurophotonics applications and in fully parallel two-photon optogenetics [3] which applies the most advanced optical tools for exploring neuro-scientific challenges. Beyond the research laboratories, efficient light shaping is also desirable for applications such as laser machining, lithography and future laser-based digital cinemas to name a few. These diverse applications all require light to be shaped in a plurality of ways [4]. For example, the illuminated optical window of spatial light modulators, used for both optics research and consumer display projectors, have a rectangular form factor. A variety of shapes bounded by steep edges and particular point spread functions are desirable in laser cutting and engraving. In two-photon optogenetics [5], it is a key aim to selectively illuminate intricate patterns of dendrites or axons within neurons, preferably with minimal loss of light and maintaining speckle-free light excitations even within turbid media.

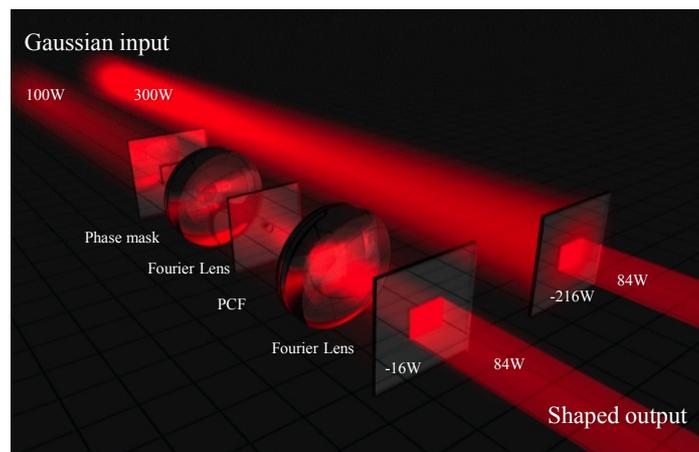


Figure 1: GPC efficiently transforms an incident Gaussian beam into a bright shaped output using only simple binary spatial phase modulation. For comparison an amplitude masking configuration is shown besides a GPC Light Shaper to illustrate the significant difference in energy utilization when aiming for the same shaped output. (Figure adapted from [6])

Laser sources typically exhibit a Gaussian intensity profile. Shaping such a beam with the commonly applied hard truncation is inherently highly inefficient. It is well known that more than two thirds of an incident power will be lost when homogeneously illuminating a rectangular aperture with an expanded Gaussian beam [6-8]. To complicate things, this lost light power will inherently contribute to device heating that can either shorten device lifespan or require additional power for active cooling. Besides the obvious disadvantages of light inefficiency, the high price tag of advanced laser sources, such as femtosecond lasers or supercontinuum sources, used for multi-photon excitation, multi-spectral biophotonics and other state-of-the-art experiments, demands efficient use of the available photons

GPC (for Generalized Phase Contrast) belongs to the class of non-absorbing common-path architectures [9]. A phase-only aperture directly representing the desired output intensity is mapped through the interference of its high and phase-shifted low spatial frequencies. This is achieved by phase shifting the lower spatial frequencies through a binary phase contrast filter (PCF) at the optical Fourier plane (cp. Fig. 1). GPC can thus be implemented with binary phase plates that are inherently simple to mass-produce with standard foundry processes common for silicon devices or microelectronics. The use of a one-to-one mapping geometry in GPC avoids dispersion effects which makes it advantageous for use with multiple wavelengths [10,11], spectrally broad light sources or for temporal focusing which can effectively confine light along the axial direction. Recently GPC also demonstrated its inherent adaptivity for boosting computer holographic reconstructions encoded on reconfigurable spatial light modulators [12].

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Remote axial positioning of temporally focused holographic patterns

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Two-photon excitation with temporally focused pulses can be combined with phase-modulation approaches, such as computer-generated holography and generalized phase contrast, to efficiently distribute light into two-dimensional, axially confined, user-defined patterns [1,2]. These light patterns are extremely robust against the effects of scattering and can propagate through hundreds of microns into brain tissue without significant degradation [3,4]. However, thus far these approaches were limited to the generation of patterns focused at the objective focal plane.

For temporally focused Gaussian beams, it was shown both theoretically and experimentally that the temporal focal plane can be axially shifted by applying a quadratic spectral phase to the incident beam (Group Velocity Dispersion, GVD) [5]. However, the case for complex wavefronts is not straightforward. Here, present an analytical, numerical and experimental study of this phenomenon and the conditions that enable remote axial control of temporally focused holographic patterns.

Remote axial displacement of holographic patterns enables coupling of holographic illumination with a second imaging or stimulation channel, providing independent control of their respective focal planes, as well as remote volume scanning.

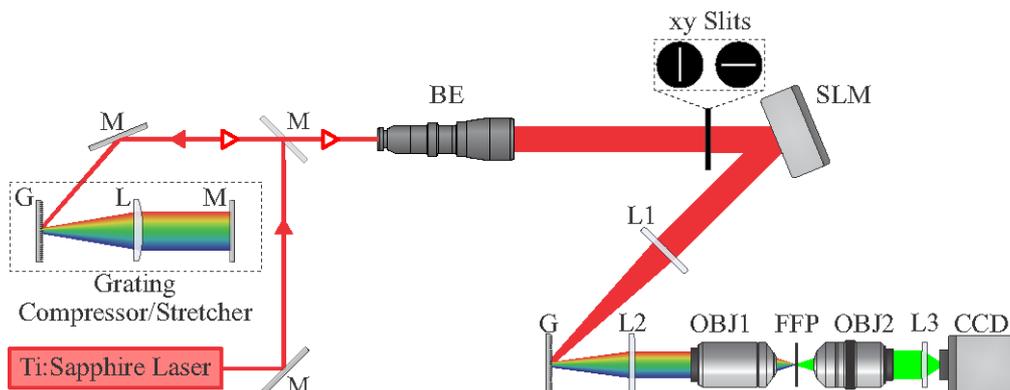


Figure 1: Schematic of an experimental setup for remote axial displacement of temporally focused holographic patterns. Laser beam from a Ti:Sapphire laser is passed through a grating compressor/stretcher where GVD is applied. The beam then impinges on an SLM and focused into the TF setup, constituted of a diffraction grating, G, and an imaging system. L: Lens, M: Mirror, BE: Beam Expander, OBJ: microscope Objectives, FFP: Front Focal Plane.

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