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Synthetic holography with spatial light modulators for biophotonics applications

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Optical wavefront shaping with spatial light modulators (SLMs), such as deformable mirrors, digital micro-mirror devices or liquid crystal (LC) panels, has become a powerful tool in Biophotonics [1]. LC-SLMs are particularly flexible, since they contain millions of individually addressable pixels to create high-resolution phase-actuation patterns. The phase-delay introduced by a pixel is determined by the tilting angle of the birefringent liquid crystal molecules which is set by applying a voltage to the pixel. Binary LC-panels, which only have 2 phase delay values corresponding to 0 and π , can typically operate much faster (at kHz refresh rate) than the types of LC-SLMs which allow 8-bit or more in phase level depth. The latter, however, support more possible settings and have higher diffraction efficiency.

A typical application of phase-masks on an SLM is the realization of programmable gratings or Fresnel lenses, for instance to create “holographic optical tweezers” comprising of one or more steerable laser spots. Optical tweezers have become a widespread tool in biomedical research, since they allow one to capture and move microscopic objects in a liquid environment in a precise and non-invasive way. Moreover, optical traps provide a means to quantify mechanical forces at work in cell biology *in situ*, for instance between motor molecules and filaments. A

disadvantage of optical tweezers is the fact that the achievable force range is usually limited to the pico-Newton regime, as the optical power required to go higher in force would induce considerable heating by absorption. Combining optical trapping with another static, but strong force field – such as a MHz ultrasound wave in a microfluidic chamber – one can levitate heavy particles while still profiting from the full steering capability of holographic tweezers [2], cf. Fig. 1.

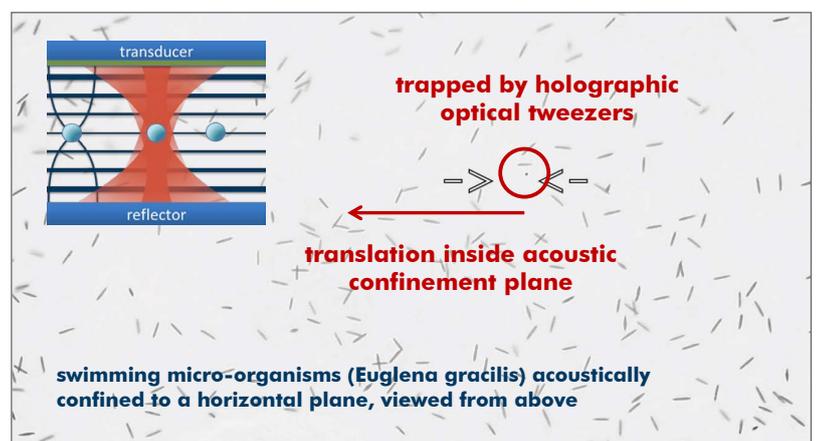


Fig. 1: Interactively steerable holographic optical tweezers created by wavefront shaping with a spatial light modulator with an ultra-large manipulation volume: In order to be able to manipulate large (length $70\ \mu\text{m}$) actively swimming micro-organisms, an auxiliary acoustic standing wave is used.

But the use of wavefront shaping is not restricted to applications in optical micromanipulation: An SLM can also be integrated into optical imaging systems, making the microscope or imaging system programmable and adaptable with respect to the needs of specific samples. In the synthetic holography approach, one *calculates* the holographic phase masks and sends them to the SLM which acts as a reconfigurable phase mask.

Placing the SLM in a Fourier plane with respect to the sample, one can emulate various microscopy techniques with the SLM acting as a programmable Fourier filter and rapidly toggle between them (cf. Fig. 2). Wavefront shaping with SLMs also enables one to reconstruct intensity patterns in the far-field or in a specified plane (cf. Fig. 3a), for example with the goal of targeting structures for optogenetic stimulation of neurons in 3D in the depth of brain tissue.

A particular strength of the synthetic holography approach is the possibility to *multiplex*, which means that one can ‘pack’ several tasks into one computer-generated hologram [3]. One can, for instance, record microscopic images that contain sub-images belonging to different imaging modalities, or from different depths inside the sample, or with different parameter settings.

It is even possible to read out several holograms for specific illumination colours from the same phase mask: Simultaneously incident illumination wavelengths will suffer different phase retardation from the same voltage pattern applied to the display, because of the difference in optical thickness of the LC layer and due to its colour-dispersion. The freedom of being able to add multiples of 2π without changing the effect of a diffractive optical element gives one the necessary degrees of freedom to accommodate several holograms, for instance for RGB-colour projection of a holographic image (cf. Fig. 3b). In this way, one may also incorporate optical imaging (in the visible) and optical trapping (in the near-infrared) in one and the same phase mask pattern.

Wavefront shaping with SLMs has become indispensable in many fields of applied optics, and will continue to make optical imaging and trapping programmable and adaptable with ever-increasing speed and control of detail.

[1] Maurer, C., Jesacher, A., Bernet, S. and Ritsch-Marte, M., 2011. *What spatial light modulators can do for optical microscopy*, Laser & Photonics Reviews, 5(1), pp.81-101.

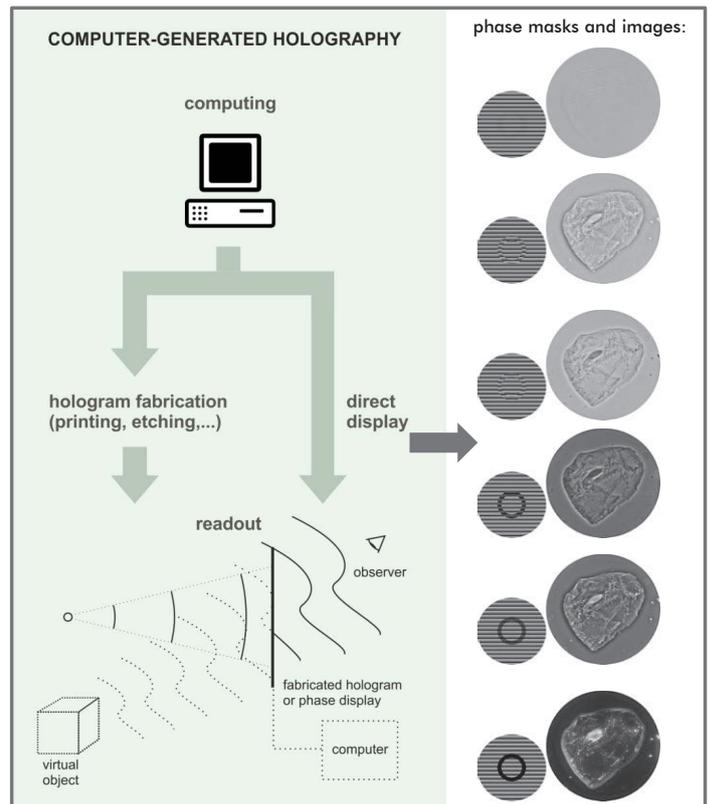


Fig. 2: In the synthetic holography approach phase patterns performing a desired task are calculated and sent to the SLM which acts as a reconfigurable phase mask. In this way, various microscopy techniques can be emulated by simply changing the phase mask, to change to a different imaging modality or to customize the parameter settings to match a given sample - without any adjustment of hardware components. In the example on the right, a thin phase object (an unstained epithelial cell) is imaged by phase masks inspired by Zernike phase contrast. The varying modulation depth of the saw-tooth grating inside the ring, as well as its relative phase shift of the gratings inside and outside the ring, influence the appearance of the image substantially. One can go from an un-contrasted bright-field image (top), to an optimized phase-contrast image, to a dark-field image (bottom) [3].

[2] Thalhammer, G., Steiger, R., Meinschad, M., Hill, M., Bernet, S. and Ritsch-Marte, M., 2011. *Combined acoustic and optical trapping*, Biomedical optics express, 2(10), pp.2859-2870.

[3] Jesacher, A. and Ritsch-Marte, M., 2016. *Synthetic holography in microscopy: opportunities arising from advanced wavefront shaping*, Contemporary Physics, 57(1), pp.46-59.

a)

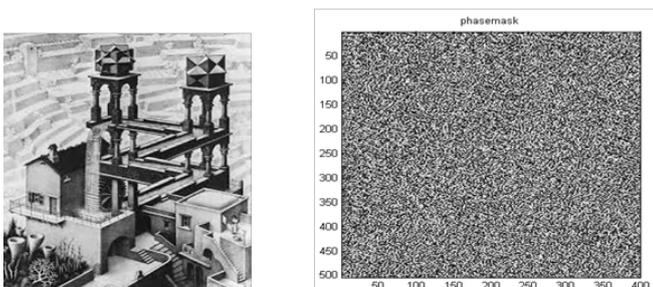
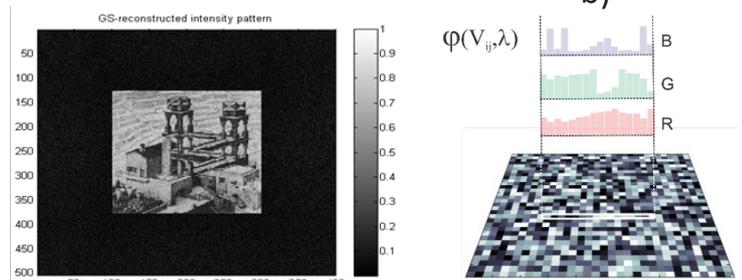


Fig. 3: Iterative algorithms are used to find a phase mask which reconstructs a desired intensity distribution in the far-field or in a specific distance from the phase mask. Fig. a) shows the image to be reconstructed (left), the phase mask calculated by a Gerchberg-Saxton search algorithm (middle) and the simulated reconstruction in the far-field (right). b) A given voltage pattern applied

b)



to the SLM panel is read out differently by different wavelengths, due to differences in the optical path length as well as colour dispersion in the LC layer. This may be utilized to multiplex several holograms into one pattern, e.g. for multi-colour holographic image reconstruction, or for combined optical trapping (by a near-infrared laser beam) with SLM-augmented imaging (in the visible).