

Research Highlights

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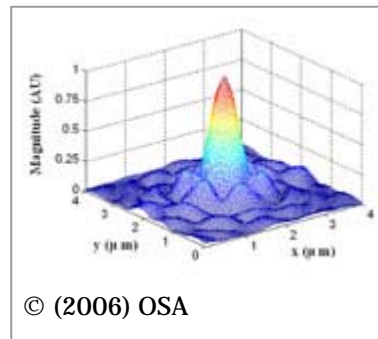
Subject Category: [Imaging and sensing](#)

Holographic microscopy: Challenging the Rayleigh limit

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Superposition of off-axis holograms drives three-dimensional imaging to subdiffraction limit

Holographic microscopy has now been upgraded to subdiffraction-limit imaging. Although such super resolution can already be achieved in two dimensions, using a series of 2D images to generate information in three dimensions can be extremely time-consuming. Scanning holography — in which a phase-encoding excitation pattern is scanned over a 2D raster — provides full 3D information, but the resolution of current holographic techniques is not as competitive as their 2D counterparts. With a simple modification to the standard scanning holography setup, G. Indebetouw *et al.*¹ were able to double the resolution possible.



In conventional scanning holography, the hologram is reconstructed from the interference between a spherical and a plane wave in the entrance pupil objective. The cutoff of the spatial-frequency spectrum, or Rayleigh limit, which determines the hologram resolution, is determined by the numerical aperture of the objective and the wavelength of the radiation. However, by combining several off-axis holograms with different spatial-frequency shifts it is possible to construct an image with a resolution beyond this apparent limit. To realize this mechanism, the team introduced a wedge prism into the scanning holographic microscopy setup. This prism could be rotated to cover various areas of the object's spatial-frequency spectrum. Preliminary experimental results for an objective with a theoretical Rayleigh limit of 0.9 μm showed that a resolution of 0.6 μm could be achieved. The researchers then combined three off-axis holograms of fluorescent beads and found a transverse resolution gain of nearly a factor of two. With more than three holograms, the resolution could be improved even further.

REFERENCES

1. Indebetouw, G., Tada, Y., Rosen, J. & Brooker, G. Scanning holographic microscopy with resolution exceeding the Rayleigh limit of the objective by superposition of off-axis holograms. *Appl. Opt.* (2006). doc. ID 74610

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