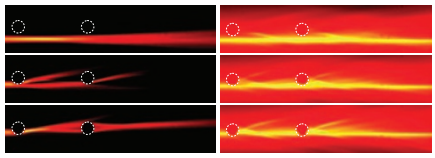
MICROSCOPY

Self-healing light beams

The self-reconstructing properties of Bessel beams provide healing benefits in highly scattering media.

The scattering of light in a scanning-microscope illumination beam has been a longstanding problem when imaging biological samples. But the magnitude of the problem is masked when a single objective is used for illumination and detection. In light-sheet microscopy, however, the use of a dedicated 'detection' objective orthogonal to the light sheet illuminating the focal plane unmasks artifacts caused by scattering of light sheets with Gaussian axial profiles. Several years ago, when Ernst Stelzer was developing his light-sheet microscopy technique, Alexander Rohrbach—then a postdoc in Stelzer's laboratory—thought that using a scanned Bessel beam instead of a cylindrically focused light sheet could minimize scattering.

In 2006, when Rohrbach started his own lab, he still had the idea of building a better light-sheet microscope using Bessel beams. "Everything started with this idea and computer simulations," says Rohrbach. The simulations showed that unlike in a conventional Gaussian beam, which carries nearly all its energy in a single central peak, the complex ring system surrounding the sharp central peak in a Bessel beam contains a large fraction of the total beam energy and should help replace scattered light lost from the central peak, thus reconstructing the beam.



Self-reconstructing beams. A Gaussian beam (left) is scattered by glass spheres (dashed circles) embedded in fluorescing gel but a Bessel beam (right) is not. Image adapted from *Nature Photonics*.

Rohrbach's graduate student Florian Fahrbach then built a microscope to verify the simulation results. They showed that Bessel beams exhibited 'self-healing' abilities via self-reconstruction just as the simulations predicted. They demonstrated this for beams interacting with large scattering spheres, clusters of small spheres and highly scattering human skin.

As with most techniques, there are tradeoffs. "The ring system is absolutely necessary but also annoying because it excites fluorophores above and below the focal plane," says Rohrbach. He and Fahrbach are hard at work on solutions, and these self-healing light beams are certain to appear in many applications in the near future.

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RESEARCH PAPERS

Fahrbach, F.O. *et al.* Microscopy with self-reconstructing beams. *Nat. Photonics* advance online publication (12 September 2010).