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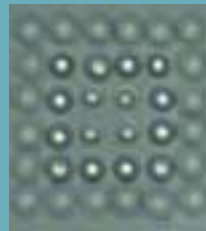
METROLOGY

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FABRICATION

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PRODUCT GUIDE

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EDITORIAL

Editor Oliver Graydon
Tel: +44 (0)117 930 1015
oliver.graydon@iop.org

Technology editor Jacqueline Hewett
Tel: +44 (0)117 930 1194
jacqueline.hewett@iop.org

Reporter James Tyrrell
Tel: +44 (0)117 930 1256
james.tyrrell@iop.org

Production editors Lucy Farrar, Clare Sturges
Technical illustrator Alison Tovey

EUROPE/ROW SALES

Advertising sales manager Rob Fisher
Tel: +44 (0)117 930 1260
robert.fisher@iop.org

Key accounts manager Simon Allardice
Tel: +44 (0)117 930 1284
simon.allardice@iop.org

Key accounts manager Adrian Chance
Tel: +44 (0)117 930 1193
adrian.chance@iop.org

Sales executives Cadi Jones, Jo Hook
Tel: +44 (0) 117 930 1090/1028
cadi.jones@iop.org/jo.hook@iop.org

US SALES OFFICE

IOP Publishing Inc, Suite 929, 150 South
Independence Mall West, Philadelphia PA 19106, USA
Tel: +1 215 627 0880
Fax: +1 215 627 0879

ADVERTISING PRODUCTION

Advertising production coordinator Teresa Honey
Tel: +44 (0)117 930 1040
teresa.honey@iop.org

Advertising production editor Tanwen Haf

CIRCULATION AND MARKETING

Product manager Angela Peck
Tel: +44 (0)117 930 1025
angela.peck@iop.org

ART DIRECTOR

Andrew Giaquinto

PUBLISHER

Geraldine Pounsford
Tel: +44 (0)117 930 1022
geraldine.pounsford@iop.org

PUBLISHING DIRECTOR

Richard Roe

OPTO & LASER EUROPE

Dirac House, Temple Back,
Bristol BS1 6BE, UK.
Tel: +44 (0)117 929 7481
Editorial fax: +44 (0)117 925 1942
Advertising fax: +44 (0)117 930 1178
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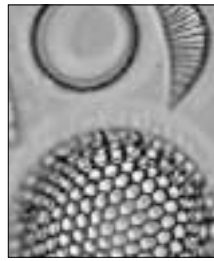
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Southall, Middlesex UB1 2DB, UK.
Tel: +44 (0)208 606 7518. Fax: +44 (0)208 606 7303.
E-mail: opto&lasereurope@iop.org

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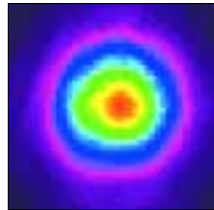
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Optical manipulation platform

Danish scientists have harnessed the power of light beams to manipulate 3D arrangements of particles at the click of a mouse button. **Rob van den Berg** reports.

A good description of Jesper Glückstad would be an architect of the microscopic world. Over the past two years, he and his co-workers Peter John Rodrigo, Vincent Daria and Ivan Perch-Nielsen have been developing machinery to manipulate micrometre-sized particles into 3D structures at the click of a mouse button. What's more, the research team, from the Risø National Laboratory in Roskilde, Denmark, do this using nothing but focused beams of light.

The team's apparatus opens up promising new possibilities in the field of materials science and biomedical research – assembling photonic crystals, seeding crystal growth or sorting cell colonies are just a few examples. When *OLE* spoke to Glückstad, he was about to leave for the US to meet with scientists from the National Institute of Health, who are keen to apply his technique to cancer and cell research.

Although the concept of the optical trapping of particles has been around for some time (Alan Ashkin and his co-workers from Bell Labs in the US demonstrated optical tweezers in the mid-eighties), Glückstad's approach allows many particles to be manipulated simultaneously in real time.

Instead of using a single laser beam to trap a particle, which has the disadvantage that it can escape if the beam is not tightly focused, the Danish team uses a pair of laser beams to move each particle. "A particle can be trapped between two counter-propagating beams that have their waists slightly separated along the optical axis," explained Glückstad.

This dual-beam approach, which Glückstad fondly refers to as an "optical elevator", has several benefits over conventional optical tweezers and traps. "Optical tweezers require objective lenses with a very high numerical aperture (NA) to generate a sufficiently strong gradient force. If such a lens is used away from the design plane, the focus is smeared out and the particle is lost," said Glückstad.

"Also, when working with live cells, it is better not to focus too tightly in order not to damage the organelles inside the cell," he continued. "We can work with much simpler optics and can also move them [the particles] over a much larger depth, even out of view of the microscope lens."

By changing the relative powers of two opposing, orthogonally polarized laser beams, Glückstad and his colleagues can vary the axial position of a trapped particle by tens of micrometres. And, thanks to a highly efficient phase- and polarization encoding scheme, they can generate many simultaneous elevators to trap and manipulate up to 80 particles with less than 1 W of near-infrared laser light.

The power of polarization

The apparatus works as follows: an expanded TEM₀₀ mode from a Ti:sapphire laser at 830 nm illuminates a spatial light modulator (SLM) and becomes encoded with a 2D phase-distribution. "This phase distribution can be represented by a spatial pattern of up to 256 levels on a computer monitor, where each grey level represents a different phase-delay between 0 and 2 pi," explained Glückstad.

"This is then projected onto the surface of a spatial polarization monitor (SPM), which acts as a variable waveplate," he continued, "and endows each beam with varying components of s-polarized and p-polarized light." Finally, a polarizing beamsplitter separates the orthogonal (s and p) components of the beams before they are directed to the sample via microscope objectives.

The beauty of the process is that it all occurs rapidly and with almost no optical loss. The system is aligned in such a way that the s and p beams are focused along a common optical axis, but have a slight axial separation in order to perform the trapping. Controlling the relative intensities of the two beams adjusts the position at which a particle finds its equilibrium. In principle, this could be done with any two counter-propagating beams, but, as Glückstad explained: "We use orthogonal polarizations because the beams should not show any interference. We do not want them to see each other."

User-friendly system

Glückstad's team have developed the system to be as user-friendly as possible. "There is no need to calculate anything – [to operate the traps] one just images the particles on the screen and superimposes them with a cursor. It is like clicking and dragging icons on your PC," said Glückstad. "With



The Danish team has used its system to organize tiny 2–3 μm-diameter patterns, including a pyramid. Particles manipulated to form the different heights (middle right, bottom). Schematic of the particle

"We have made a wonderful tool. Now we want to apply it."

Jesper Glückstad

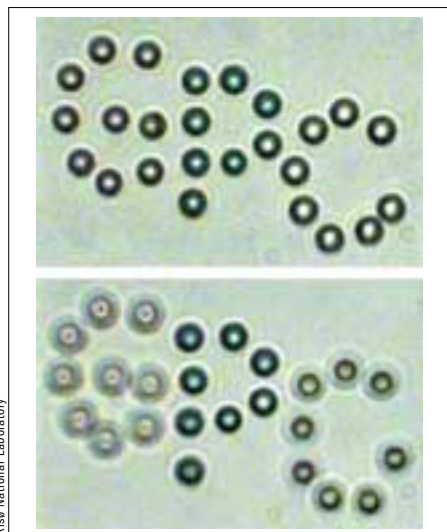
the left mouse button you can move them around in the plane, and with the right button you can determine the depth. The speed with which we can move the particles around is tremendous – up to 25 μm/s." A further benefit of the system is that it offers a 3D view of the location of the particles while they are being manipulated. "This capability is unique, since it allows us full visualization and precise position and velocity control," commented Glückstad.

To demonstrate the potential of the technique, the team have assembled 2–3 μm-diameter polystyrene and silica spheres into

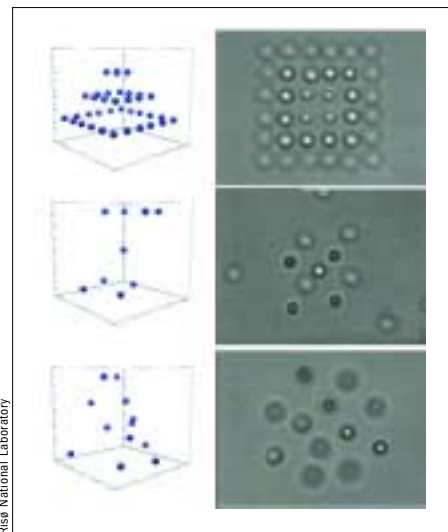
ays tricks with particles



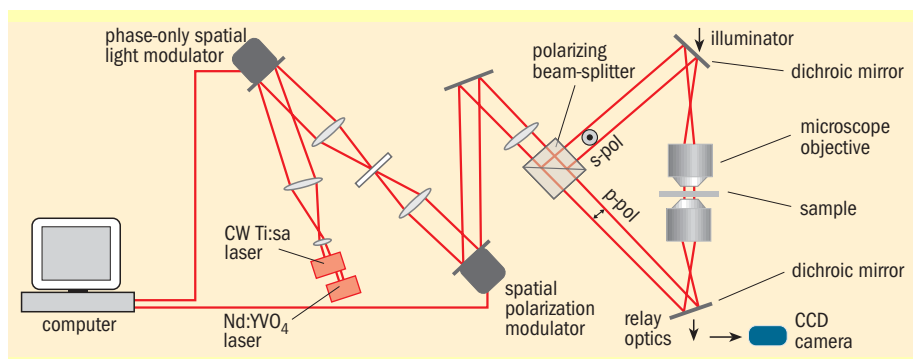
meter polystyrene and silicon dioxide spheres (far right) into 3D letters GPC in a single plane (middle right, top) and at three the manipulation system (right).



Risø National Laboratory



Risø National Laboratory



all kinds of 3D arrangements (see above), such as a pyramid and a series of letters (*Applied Physics Letters*, 14 February 2005).

Real-world applications

Significantly, the wavelength of 830 nm is not absorbed by living tissue, so living biological matter can be manipulated without any risk of damage. In an upcoming paper in the journal *FEMS Microbiology Letters*, the Danish team also reports the first “real-world” application of the technique: a study of the growth of different yeast species.

“It has been found that, when mixing two types of yeast – *Scerevisiae* (S101) and *Huvarum* – the strong S101 cells surround the weaker uvarum cells and stop them from growing. With our traps, we were able to controllably surround the individual cells of one species with those of another [and analyse the affect on growth],” said Glückstad. “We found that the average generation time of surrounded cells was 15% longer than that of non-surrounded cells, thereby showing that confinement inhibits growth.”

In a further paper in the online journal *Optics Express*, Glückstad and his team describe experiments with low-NA optics and a long working-distance of more than 10 mm. The result is not only a much wider manipulation region, but also a larger field-of-view for imaging.

“We can observe the trapped particles simultaneously from the top and from the side. Such visual data could, in principle, be obtained using a confocal microscope, but we can now capture images from two orthogonal planes simultaneously and in realtime,” said Glückstad. “This enables us to calibrate and fine-tune the counter-propagating beams and observe the dynamics of particles that lie well outside the focal plane of an ordinary top-view microscope.”

The long working-distance also makes it possible to manipulate particles in devices like microfluidic (lab-on-a-chip) systems that are too cumbersome to fit into a microscope equipped with high-NA immersion objectives.

In the near future, Glückstad hopes to do experiments with stem cells. These are

embryonic cells that can develop in different ways depending on the cells that surround them. By surrounding them selectively with specific cell types, he would like to check their development in a controlled way. He also wants to investigate cancer cells and is currently drafting several project proposals with scientists from the US.

Apart from finding promising applications for his technique, Glückstad has worked hard to design and build a commercial system, and has been talking to manufacturers who are interested in marketing an upgraded version of the set-up.

“It [the new version] will be much simpler, more robust and faster than our current one. There have been new developments in SLM technology,” said Glückstad. “The ones we are using are based on nematic liquid crystals, which are relatively slow and offer a refresh rate of some 5 fps. Nowadays, this can be done several orders of magnitude faster.”

Rob van den Berg is a freelance science and technology journalist based in the Netherlands.