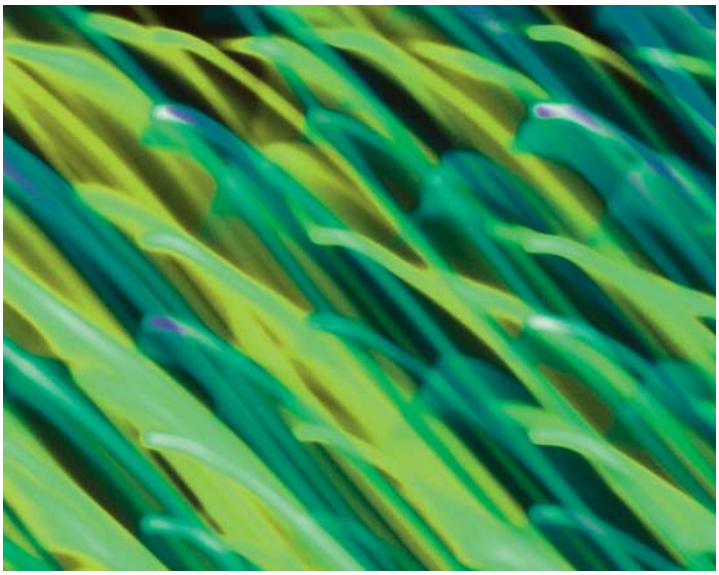
TECHNIQUES

# Bright future for GFP



One of the most useful cell-biological tools to have been introduced in the past decade is green fluorescent protein (GFP). And it seems that its star is still rising, as Shifang Zhang, Charles Ma and Martin Chalfie describe in *Cell*.

In its simplest form, the fluorescence produced by GFP is used to reveal where a protein of interest is expressed, by fusing the coding sequence of the protein to that of GFP. More recently, it's been shown that GFP can also be used to monitor protein–protein interactions, *in vitro*, in bacteria or in cultured cells. To achieve this, GFP is split into two polypeptides, each of which is attached to a protein-interaction domain, or to a target protein. If the two protein-interaction domains — or the two target proteins — bind to each other, then the two GFP polypeptides are brought together, and the GFP fluoresces.

Chalfie and colleagues have now demonstrated numerous other uses for what they call 'reconstituted GFP' (recGFP). First they showed that recGFP — consisting of two GFP segments, each attached to a protein-interaction domain — can be generated *in vivo*, specifically in *Caenorhabditis elegans*. They also found that cyan fluorescent protein and yellow fluorescent protein can each be split up and reconstituted in the same way in *C. elegans*.

The authors then investigated whether the recGFP system could be used to identify cells that co-express two different genes. In this case, they already knew that the *C. elegans unc-24* gene is expressed in the six touch-receptor neurons and some cells in the ventral cord; *mec-2*, by contrast, is expressed only in the touch receptors. Expressing one GFP polypeptide from the *unc-24* promoter and the other from *mec-2* reconstituted GFP only in the touch receptors, which confirmed that these two *C. elegans* genes are co-expressed only in these neurons. This is a proof of principle; the technique could now be used to identify cells in which genes with previously unknown expression patterns are co-expressed.

The recGFP system could also be used to identify cells that express a particular gene. Chalfie and colleagues chose to look at the *sto-6* gene, the expression pattern of which was unknown. They found that this gene, fused to the regular GFP sequence, was switched on in many different motor neurons in the ventral cord. To find out exactly which neurons, they used recGFP: by expressing one GFP polypeptide from the *sto-6* promoter and the other polypeptide from the promoters of various genes for which the expression patterns were known, the authors tracked down the expression of *sto-6* to the excitatory motor neurons.

Chalfie and colleagues tested other uses for recGFP, such as labelling cell constituents in a restricted group of cells *in vivo*. And they propose further possibilities — demonstrating cell fusion or viral infection, for instance, in *C. elegans* and beyond. The future of GFP looks bright indeed.

*Amanda Tromans, Senior Editor,* Nature

## References and links

**ORIGINAL RESEARCH PAPER** Zhang, S., Ma, C. & Chalfie, M. Combinatorial marking of cells and organelles with reconstituted fluorescent proteins. *Cell* **119**, 137–144 (2004)

**WEB SITE**
**Martin Chalfie's laboratory:** http://www.columbia.edu/cu/biology/faculty/chalfie/