The brightest dots in the bunch

Quantum dots (QDs), fluorescent inorganic crystals <10 nm in diameter, offer several advantages over organic dyes for labeling biological samples. They resist photobleaching, and their emission spectra are narrow enough that multiple variants (excited by one wavelength) can be distinguished in a given sample. The feasibility of using QDs for biological uses is proven in two recent articles by Jyoti Jaiswal, Sanford Simon (Rockefeller University, New York, NY), and colleagues and by Xingyong Wu, Marcel Bruchez (Quantum Dot Corporation [QDC], Hayward, CA), and colleagues. “Quantum dots were offered up as a savior for protein labeling a couple of years ago,” says Simon. “But it hadn’t been shown that they could be used in vivo.”

The QDC group shows that QDs can be used to label proteins in fixed and living cells. Using two colors of IgG- and streptavidin-attached QDs, they simultaneously identified both nuclear and plasma membrane proteins with only a single filter set in fixed cells. QD emissions were four to nine times brighter than Alexa, one of the brightest organic dyes.

Simon’s group extended the usage to long-term labeling of living cells. QDs covered with a stable chemical coat were taken up by endocytosis or were linked to antibodies to label specific membrane proteins. As the label did not affect cell growth or development, the stability of QD fluorescence allowed the authors to follow developing amoebae over several hours and mammalian cells for over 12 d. Simon looks forward to using QD technology to examine protein dynamics during vesicle fusion to the plasma membrane, a process not well suited to the rapidly photobleached GFP.

In the future, nonendocytic organelles may be specifically labeled by conjugating QDs to a signal peptide that will target them to the desired organelle. Labeling of specific nonmembrane-associated proteins inside a living cell may be done by conjugation of the QDs to protein-specific ligands or antibodies. However, for now, those labels will have to be delivered into the cell using the same invasive techniques as for conventional membrane-impermeant dyes.

References: Jaiswal, J., et al. 2002. Nat. Biotech. 10.1038/nbt767.
Wu, X., et al. 2002. Nat. Biotech. 10.1038/nbt764.

Regulated exocytosis has its own organelle, according to Barbara Borgonovo, Jacopo Meldolesi (Vita-Salute San Raffaele University, Milan, Italy), and colleagues. Various signals elicit exocytosis. The best-studied example is regulated secretion, in which a specific cargo is delivered outside the cell. But regulated exocytosis can occur in nonsecretory cells, begging the question of which organelle in these cells responds to the stimulus. Recently, lysosomes were shown to fuse to the PM in response to damage-induced calcium signals, but they no longer appear to be the only organelle in the arsenal.

By comparing the PM of living cells before and after stimulation, Meldolesi’s group has now identified desmoyokin (dA), a marker for regulated exocytosis that appears on the PM in response to increases in intracellular calcium. dA was found in a set of small vesicles within 0.5 μm of the PM that fused rapidly in secretory and nonsecretory cells. The vesicles lacked markers for known organelles, including endosomes, trans-Golgi, and lysosomes, and are thus a distinct class.

The group named the new organelles enlargosomes. “The name we gave them shows what we have in mind [for their function],” says Meldolesi. He thinks the organelles may provide cells with a means to increase PM surface area during wound healing without releasing hydrolytic enzymes. More enlargosomes were found in differentiating cells, which are also expected to be expanding. Enlargosomes may favor a lasting increase in cell size, as they were recycled more slowly than vesicles involved in regulated secretion.

Reference: Borgonovo, B., et al. 2002. Nat. Cell Biol. 4:955–963.