

TOOLBOX

New method exposes structures inside 'rainbow' of brain cells

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4 JANUARY 2019

Molecules from alpacas may enable scientists to identify cell types in the brain while also revealing their interior structures¹. The method may help researchers better visualize how the brain is wired in autism.

Researchers often stain tissue with fluorescent antibodies to certain proteins so they can identify cell types. They can also use electron microscopy to view the tissue's internal structure.

But the two techniques can't be used on the same sample. The former requires chemicals that often break down cells. Electron microscopy involves thinly slicing the sample, and staining the

numerous slices afterwards is prohibitively laborious.

Scientists have **engineered brain cells** to produce fluorescent proteins or made tags that are visible in an electron microscope. But these methods can label only about three cell types per sample. The new technique lets researchers tag tissue with up to 10 markers.

The researchers stained sections of mouse brain using fluorescent ‘nanobodies’ — antibody fragments typically derived from camelids, the mammalian family that includes camels, alpacas and llamas. (The nanobodies they used came from alpacas.) Because of their small size, nanobodies can infiltrate tissue without it needing to be pretreated with harsh chemicals.

Cell snapshots:

The team created images of the stained tissue with a fluorescent microscope. They then sliced the samples into 50-nanometer-thick segments and scanned those with an electron microscope. Using computer software, they aligned both sets of images based on landmarks such as blood vessels.

The resulting images highlight four cell types, including star-shaped **support cells called astrocytes** and immune cells called **microglia**, each labeled with different colors. The fluorescent nanobodies penetrate more than 100 micrometers into the block of tissue. By contrast, antibody markers stain only the surface.

The images show organelles such as **mitochondria** (the cell’s energy producers) and microtubule bundles (the cell’s structural skeleton). The researchers could also make out the long filaments that extend from neurons, and the vesicles at the end of them that hold **chemical messengers**.

The nanobody method could reveal differences in cell structure and the distribution of cell types in the autism brain, says lead investigator **Jeff Lichtman**, professor of molecular and cellular biology at Harvard University. His team described the technique in November in *Nature Methods*.

The ultimate goal, Lichtman says, is to stain a sample with hundreds of colors by developing more nanobodies and combining fluorescent dyes.

REFERENCES:

1. Fang T. et al. *Nat. Methods* **15**, 1029-1032 (2018) [PubMed](#)