

Exploring 3-D Space

After a 20-year wait, a team of scientists is building an atlas to map individual brain neurons the way astronomers identify stars in the sky.

TREATING GENE EXPRESSION PATTERNS AS A SEARCHABLE CONSTELLATION of stars. The idea was too far-fetched to tackle two decades ago, when Chris Q. Doe was a postdoctoral fellow. He wanted to build an atlas showing the three-dimensional location of transcription factors involved in generating the fruit fly’s central nervous system (CNS). Other atlases, however, covered simpler one- and two-dimensional patterns.



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CHRIS DOE

As proteins responsible for initiating gene expression, the many transcription factors in a fruit fly CNS correspond to different types of neurons. Making sense of these patterns could help scientists discover how brain cells develop and go on to form complex circuits.

Doe, now an HHMI investigator at the University of Oregon, gives credit for the idea of creating an atlas to his former postdoctoral adviser, HHMI investigator Matthew P. Scott, who is now at Stanford University. Scott had developed an atlas of *Hox* gene expression for *Drosophila*. He and his colleagues discovered that *Hox* genes, which determine where limbs and segments will form in the developing embryo, are expressed in a one-dimensional pattern. When Doe struggled to make sense of his three-dimensional data, he recalls Scott saying: “Just build an atlas.”

Easier said than done. “I remember thinking it would be impossible—and that was with only a handful of CNS genes identified,” says Doe. He dropped the idea as technologically unfeasible. The seed, however, was planted.

Now, 20 years and more than 200 transcription-factor genes later, Doe and his postdoc Michael Layden are on the verge of seeing such an atlas become a reality, thanks to a collaboration with computer scientist Eugene W. Myers, a group leader at HHMI’s Janelia Farm Research Campus. Doe and Myers met at a neuroscience conference held at the Ashburn, Virginia, campus in March. “We’ve been working hard at this ever since,” says Doe. “The methods of computational biology are now so advanced that it’s conceivable to make a computer atlas of the nervous system and map the expression of hundreds of genes.”

And if that atlas were searchable, he notes, it would potentially allow scientists to solve two important mysteries in neurobiology: how a stem cell becomes a particular type of neuron, and how neurons come together to

Illustration: Jon Han Photo: John Clark



form circuits that allow for all the complex processes governed by the brain. “Our best hypothesis is that neurons that make up a circuit share a transcription-factor code,” says Doe. “Many labs are looking to identify these circuits. We’re hoping our atlas will help them determine which transcription factors those neurons have in common.”

Then researchers could work backward to figure out which genes direct progenitor cells to differentiate into particular types of neurons, says Stephen Crews at the University of North Carolina at Chapel Hill. Crews has created a two-dimensional atlas of gene expression for more than a hundred genes involved in the development of *Drosophila* midline CNS. Because transcription factors are largely conserved throughout evolution, Crews expects the three-dimensional fly atlas will help in understanding development of the nervous systems of higher animals. “Having a searchable atlas of hundreds of genes that all labs could have access to would really be a huge step forward,” he says.

To create the first draft of the atlas, Doe and Layden used fluorescent antibodies to label

neuron-specific transcription factors in late-developmental *Drosophila* embryos. Then, using confocal microscopy, they produced two-dimensional images—optical “slices”—of the resulting three-dimensional patterns of fluorescence. Doe and Layden are creating stacks of these slices for each of the more than 200 transcription-factor genes identified to date.

Myers and his team take these stacks and produce a computer-generated model for each gene that resembles a constellation—glowing points of color in a black three-dimensional space. The last step in building the atlas involves overlaying these patterns on one another. To do this, Myers’ team is writing software that aids in the recognition of the patterns created by the glowing nuclei of each neuron. “We’re learning to identify these nuclei the way astronomers identify stars in the sky,” Myers says. “How do you find the North Star? You look for the pattern created by the stars that make up the Big Dipper. In the same way, we are looking for invariant patterns.”

Making the atlas a searchable one will be the next challenge, as current search technology is limited to one dimension.



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MICHAEL LAYDEN

“We can easily do a Google search using the name of a person,” says Doe. “What we want to do would be like searching Google for the shape of a person’s head.” According to Myers, no one has yet accomplished this next-level search technology. “We’ll learn a lot of biology by doing this,” he says, “but we will also learn a lot about exploring and understanding three-dimensional space.” ■

— CAMILLE MOJICA REY

