THE AUTHOR FILE

Paul Tesar

Pure oligodendrocyte populations can be made without cell sorting.

As a postdoc and graduate student, Paul Tesar spent years learning how stem cells stay in a nondifferentiating, pluripotent state. Along with other members of

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Ron McKay's laboratory and another, independent team of researchers, Tesar discovered a new type of pluripotent cell, mouse epiblast stem cells. These behave more like human embryonic stem cells than conventional mouse embryonic stem cells.

But after so much time studying how to keep cells pluripotent, Tesar was ready to see what he could make from these cells. It is the differentiated products of stem cells, after all, that are most likely to be used to identify new therapies. Though familiar with neural cells, Tesar was not interested in joining the crowded race for differentiating stem cells into neurons. Instead, he focused on the brain's support system. "Glia are the other cell type," he says. "They make 90% of the brain, but everyone was studying the other 10%."

He moved from the US National Institutes of Health in Washington, DC to Case Western Reserve University in Cleveland with the goal of learning how to turn mouse epiblast cells into oligodendrocyte precursors, cells that eventually wrap neuronal axons in protective myelin sheaths. Better control over oligodendrocytes might help treat spinal cord injury, multiple sclerosis, cerebral palsy and even schizophrenia.

Geographically, the move was a homecoming: Tesar is a native of the state of Ohio and attended Case Western as an undergraduate. He now lives within five miles of where he grew up. Careerwise, however, it was a jolt. Tesar was jumping from an unusually short postdoc to an assistant professor position, and he was also entering a new field. The learning curve was steep, says Tesar. "I'm not a myelin guy. I'm a stem cell guy. We were a brand new lab and had no idea how to do functional assays."

Luckily, Case Western's Robert Miller agreed to collaborate from the beginning, sharing not only expertise but also reagents and other resources. It would be his lab members who injected the differentiated cells into myelin-deficient mouse brains to demonstrate that the cells researchers in Tesar's lab had generated really were oligodendrocyte precursors.

Long before there were cells to inject, however, Tesar had to work out a protocol for making them. Though

his appointment did not officially start until January 2010, his lab members managed to get a few experiments running in December. "I came in on Christmas morning, just to see the cells," Tesar recalls. "Right in the middle of the plate was an oligodendrocyte. It was the most beautiful cell that I'd ever seen." Tesar printed out the image and showed it to everyone in his family: his wife, his mother and his in-laws. "Even my dog got to see it," he says.

Oligodendrocytes are very distinctive cells, says Tesar, a round cell body surrounded by countless tiny projections. "People describe it as a bloodshot eyeball. It doesn't sound beautiful, but it is."

As exciting as that Christmas morning was, Tesar's goal went beyond just making one cell. He wanted to create large, pure populations quickly, without the need to separate differentiated cells or kill off undifferentiated ones. Protocols for making oligodendrocyte progenitor cells have been established, says Tesar, but these rely on robust purification rather than precise differentiation. That precludes several kinds of experiments. "If you have mixtures, you can't do anything to see how the cells are changing fate," he explains. And the more control researchers have in creating a certain cell type, the more likely it is that they will be able to develop products suitable for cell screening or transplantation. The most efficient way to achieve that control, says Tesar, "is to take it in a stepwise way and make sure each step is very pure."

Now Tesar is working to translate the rigorous protocols developed for mouse epiblast cells and reported on page 957, to human pluripotent cells. Though such translations have not been described before, Tesar believes it should prove relatively straightforward. After all, mouse epiblast stem cells maintain pluripotency through many of the same pathways as human embryonic stem cells do and even grow better in medium designed for the human cells than in medium for conventional mouse embryonic stem cells.

Still, applying the differentiation protocol to human cells will probably require the same commitment to rigorous trial and error. For example, explains Tesar, "everyone thinks oligodendrocytes need [the protein] sonic hedgehog, so many protocols say 'add a lot and hope for the best.'" Subtlety makes a far better strategy. "The most surprising aspect is how sensitive the cells are to the concentrations of the reagents," says Tesar. "Very small changes give you precise results."

Monya Baker

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Najm, F.J. *et al.* Rapid and robust generation of functional oligodendrocyte progenitor cells from epiblast stem cells. *Nat. Methods* **8**, 957–962 (2011).