Reining in pain

By Lev Osherovich, Senior Writer

University of California, San Francisco researchers have shown that neuronal precursor cells from fetal mouse brains can be transplanted into the spinal cord of adult mice to treat neuropathic pain.¹ The embryonic brain cells are being developed by **Neurona Therapeutics Inc.**, a company founded by several of the San Francisco team members.

The precursor cells come from the medial ganglionic eminence (MGE) region of the embryonic brain. During development MGE cells migrate to distant areas of the brain, form synapses with nearby neurons and mature into γ -aminobutyric acid (GABA)-producing inhibitory neurons, called GABAergic interneurons.

Now, a team led by Allan Basbaum, professor of anatomy at UCSF, has shown that transplanted MGE cells can take root in the spinal cord and adopt an inhibitory function that suppresses neuropathic pain.

"Inhibitory interneuron activity, which normally regulates outflow of pain, is decreased" in patients with neuropathic pain and models of the disease, said Basbaum.

Basbaum said there was prior evidence that inhibitory GABA activity in the spinal cord could suppress the transmission of neuropathic pain. Thus, his team set out to enhance this effect by increasing the number of inhibitory GABAergic neurons within the spinal cord.

"We asked if we could repopulate GABAergic interneurons in the spinal cord with cells from the embryonic cortex," said Basbaum. "We've demonstrated that these cells can integrate into the spinal cord circuitry and normalize GABA production."

New home

The team began by harvesting MGE cells from the brains of embryonic mice, marking them with transgenic GFP and injecting them into the spinal cords of adult mice. After one month, the cells had migrated throughout the spinal cord region near the injection site and differentiated into mature neurons expressing GABAergic markers.

Fluorescent labels showed that the transplanted neural precursors formed synapses with adjacent spinal cord neurons. MGE-derived neurons in the spinal cord became activated by both normal mechanical stimulation and by painful stimulus, as expected for interneurons with multiple synaptic connections.

Basbaum said there initially was some concern that having too many

GABAergic neurons in the spinal cord would create an overly inhibitory environment that would interfere with normal nervous system functions. However, "the transplant normalizes GABA production but doesn't put it above normal range," he noted.

Finally, the team assessed the functional effect of the transplant. In a functional assay of neuropathic pain, mice with MGE transplants had less pain avoidance behavior than nontransplanted controls. The MGE cells had no effect in a model of inflammatory pain, which is a distinct form of pain that does not involve dysregulation of GABA signaling.

Data were reported in *Neuron*.

Altogether, the findings suggest MGE transplants can dial down the intensity of sensory signals that drive neuropathic pain.

"In nerve injury, there can be persistent activation of brain circuits, leading to hyperexcitability," said Cory Nicholas, Neurona CEO and a postdoctoral fellow at UCSF. "This is a peripheral injury but the cells are acting in the spinal cord, inhibiting the pain pathways that are coming up the injured neurons into the CNS."

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—Allan Basbaum,
University of California,
San Francisco

Nervy play

A number of neurological conditions are associated with excessive stimulatory signaling in the brain and could be amenable to normalization by MGE-derived GABAergic cells, said Nicholas.

Indeed, prior work by members of the UCSF team and other groups has shown the possibility of using embryonic MGE cells for epilepsy and Parkinson's disease (PD).²⁻⁴ In all of these cases, the MGE cells were transplanted into the brains of mice rather than into the spinal cord.

Remaining hurdles include determining whether human MGE cells will behave similarly to mouse cells in cell culture and when transplanted into the spinal cord and scaling up the production of human MGE cells to therapeutically useful quantities.

Because human MGE cells would ordinarily need to be harvested from fetal brains, Nicholas said the company has developed a way to make MGE-like cells from human induced pluripotent stem (iPS) cells. The company is now testing those cells in multiple undisclosed disease models.

Cells derived from iPS cells typically undergo differentiation *in vitro* prior to transplantation. Thus, it remains to be seen whether iPS cell-derived MGE cells will migrate, engraft and function as efficiently as actual MGE cells.

Regardless of the source, Chris Parker, VP and chief commercial officer of **Cellular Dynamics International Inc.**, said getting useful quantities of human MGE cells and scaling up production would probably require identifying markers that distinguish the cells from other semi-differentiated cells of the embryonic cortex.

Cellular Dynamics markets iPS cell-derived cell lines as discovery tools.

Parker noted that one CDI product—the iCell Neuron preparation—

is a mixture of iPS cell-derived forebrain neurons that may include some MGE-like cells. He said the company might test whether iCell Neurons can behave like the UCSF team's primary MGEs in neuropathic pain.

Neurona is funded by grants from the **California Institute for Regenerative Medicine** (CIRM), and Nicholas said the company hopes to raise venture money once it demonstrates the preclinical efficacy of human MGE cells. The company has filed for a patent covering *in vitro* production of MGE cells. UCSF has filed for patents covering the use of MGE cells to treat neuropathic pain, epilepsy and neurodegeneration.

Osherovich, L. *SciBX* 5(25); doi:10.1038/scibx.2012.646
Published online June 21, 2012

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