Parkinson's double take

By Lauren Martz, Staff Writer

Emory University researchers have found that a mouse model of Parkinson's disease they developed in 2007 could be more useful than expected. A second look at the mice showed that the model recapitulates not only tremors and rigidity, but also nonmotor defects of the disease that are absent in other models.¹

Until about 10 years ago, PD was thought to be a motor disease resulting specifically from disturbances to the dopamine system—namely a loss of dopaminergic neurons in the substantia nigra pars compacta subunit of the brain, according to the **Parkinson's Disease Foundation**. Now, PD is recognized as a neurodegenerative disease affecting not only motor functioning, but also many nonmotor processes.

As a result of this understanding, the symptoms associated with PD have expanded to include anosmia, which is the inability to perceive smells, gastrointestinal disturbances, sleeping disorders, anxiety and depression. All of these symptoms can precede the motor symptoms of tremor and rigidity. These broader symptoms are associated with disruptions not only in dopamine transport, but also in the transport of other monoamines including serotonin and norepinephrine.

"Fifteen years ago, Parkinson's disease was viewed as a dopamine deficiency causing motor symptoms including hypokinesia that start late in life and progress," said Reinhard Horowski, CSO of **Axxonis Pharma AG**. "Now we know that there is much more than descending dopamine to take into account. The disease starts with serotonin and the sleeping and behavioral problems that its loss causes. Other early consequences include pain symptoms that might also be due in part to serotonin dysfunction."

Despite the better understanding of PD pathology and symptoms, the animal models used to test therapeutics have lagged behind. Classical models are still used and specifically focus on dopamine to capture the motor deficits of the disease.

In 2007, Gary Miller and colleagues at Emory published a paper in the *Journal of Neuroscience* that described a mouse model of PD that had 95% of vascular monoamine transporter 2 (SLC18A2; VMAT2) genetically depleted.² VMAT2 controls the packaging and transport of monoamines including dopamine, norepinephrine and serotonin. The mice had clas-

sical characteristics of PD pathology—progressive neurodegeneration associated with decreased striatal dopamine and dopamine transporter formation, and accumulation of α -synuclein.

Now, the team has run a more comprehensive analysis of the same PD mice. In a new paper in the same journal, the group reports that VMAT2 depletion also captures the nonmotor symptoms of PD. Miller is associate professor for the Center of Neurodegenerative Disease in the Department of Environmental and Occupational Health at the Rollins School of Public Health at Emory.

The researchers isolated brain sections from the VMAT2-deficient mice that included the striatum, cortex and hippocampus, and saw both the expected reductions in dopamine levels and also depletion of serotonin and norepinephrine levels compared with those in wild-type mice.

Animals deficient in VMAT2 also had a disrupted sense of smell in an olfactory discrimination test and delayed sleep latency and gastric emptying. Finally, the mice also showed anxiety-like behaviors in a

maze test and depressive immobility in forced swim and tail suspension tests as they reached 12–15 months of age. Depression and anxiety have previously been linked to disturbances in the serotonin system.³

Miller told *SciBX* that his team is planning to use the PD model to test new therapeutics.

Slow and steady

"We now use a cocktail of drugs for this disease, and this animal model is excellent because it can take into account all of the variables that must be considered for treatment," said Horowski. Although supplementing dopamine levels clearly helps the motor deficits of PD, such monotherapy "is no longer sufficient as more symptoms of the disease caused by different molecular changes are identified."

Axxonis is developing subcutaneous and transdermal formulations of Lisuride, a dopamine agonist. Both formulations are under EMEA review in Europe to treat PD.

In addition to providing a full spectrum of PD symptoms, Dinah Sah, VP of research in CNS and oncology at **Alnylam Pharmaceuticals Inc.**, thinks the Emory mice may have additional benefits over conventional models.

"This is a slowly progressive model, more reflective of the natural progression of Parkinson's disease," she said. Such slow progression makes the mice a more accurate model than the widely used chemical toxin models.

"The next steps are to provide more detailed temporal characterization of progression of symptoms, which would facilitate testing of therapeutic agents," Sah said.

Alnylam has a small interfering RNA targeting the α -synuclein gene in preclinical testing to treat PD. Sah said the mice provide another model with which Alnylam can test its therapeutic, complementing α -synuclein transgenic mice that are already available.

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**—Dinah Sah,
Alnylam Pharmaceuticals Inc.**

Sah did note that using the Emory model “will take more time” to evaluate therapeutics. “Some of the Parkinson’s disease symptoms in this model will take 6–12 months to develop,” she said.

Nevertheless, Axxonis’ Horowski said any potential time constraints could be overshadowed by the ease of use. “Primate models are particularly useful because they parallel human processes very nicely, but hurdles including cost and ethical issues associated with the use of monkeys are increasing the demand for better rodent representations” of PD, he said.

Dusting off more models

The fact that the Emory group discovered the added utility of their model on a second pass could prompt a closer look at other models of PD.

“We have been very focused on studying the dopamine depletion and motor symptoms that we have omitted the investigation of other equally important characteristics, such as cognitive impairments and peripheral problems, for example. The more classic or recent models of the disease could show these features as well, but they have not been characterized according to these criteria,” said Francesca Cicchetti, researcher at the **Research Centre of the Laval University Medical School (CHUQ)** and associate professor at **Laval University**. “It is difficult to say whether other models do or do not have this general monoamine disruption and resulting nonmotor impairments—perhaps they just haven’t been investigated or reported.”

Horowski also thinks the nonmotor PD symptoms exhibited by the Emory mice deserve further study. “I would love to perform more tests on the animals to determine if they have altered pain perception, which can also result from the disease,” he said. “We also want to see more of what is happening in the sleep patterns.”

“This model will need to be tested using symptomatic treatments,” said Gloria Meredith, chair of the Department of Cellular and Molecular Pharmacology at the Chicago Medical School of the **Rosalind Franklin**

University of Medicine and Science. “It is easy to look for the hallmark symptoms of Parkinson’s disease, but unless they can be corrected using the present-day symptomatic treatments for patients with the disease, it might not be a good testing platform for new treatments.”

Meredith added that although the nonmotor PD symptoms were well described in the paper, she wanted to see more detailed data on the motor symptoms displayed by the mice, including measures of coordination. She also wanted to see a quantitative analysis of changes in monoamine levels.

Miller told *SciBX* that Emory has not patented the model, but he said the **Babraham Institute**, which supplied the original VMAT2 knockout mice in the first paper, has IP that can be negotiated through Emory. He said the university is also interested in industry partnerships to perform studies on PD therapeutics using the model.

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