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Parkinson's Disease Mechanism Discovered

Howard Hughes Medical Institute researchers have pinpointed defects in a critical cellular pathway that can lead to the death of dopamine-producing nerve cells and ultimately symptoms of Parkinson's disease. They have also used several animal models of the disease to identify a new way to rescue dying neurons.

According to the researchers, the findings offer a promising opportunity for developing new drugs to treat the underlying causes of Parkinson's disease and related neurodegenerative disorders.

"These findings are exciting because they tell us we have a platform for discovering new therapeutic strategies and for speeding the process of discovering treatments for these disorders."

— Susan L. Lindquist

The research team, which included Howard Hughes Medical Institute investigators Susan L. Lindquist and Nancy M. Bonini, published their findings on June 22, 2006, in *Science Express*, which provides electronic publication of selected *Science* papers in advance of print. Lindquist is at the Whitehead Institute for Biomedical Research and Bonini is at the University of Pennsylvania. Antony Cooper of the University of Missouri-Kansas City and Aaron Gitler, who is in Lindquist's laboratory, were co-lead authors on the paper. Other co-authors were from Purdue University, the University of Alabama, Medical College of Georgia and New York University.

The researchers' began their experiments seeking to clarify the role of the protein alpha-synuclein in Parkinson's disease. It had long been known that abnormalities in alpha-synuclein could cause a lethal buildup of the protein in neurons. Researchers also knew that accumulation of alpha-synuclein caused neurodegeneration in animal models of Parkinson's disease, but little was known about alpha-synuclein's normal cellular function or how it contributed to disease. One major problem facing researchers, Lindquist said, was that alpha-synuclein accumulation causes a range of abnormalities, and it was not

possible to sort out which were causes and which were effects in Parkinson's disease pathology.

However, Lindquist's team developed a technique to switch on overproduction of alpha-synuclein in yeast, so they could determine which abnormalities arose earliest in the pathological process. Those experiments by Cooper revealed that an important early defect affected the machinery that transports proteins between two major cellular organelles — the endoplasmic reticulum (ER) and the Golgi apparatus. The endoplasmic reticulum is the site of protein production, and the Golgi apparatus is the cell's post office, which modifies, sorts and adds the molecular addresses that designate the specific destinations in the cell where proteins are needed.

Lindquist and her colleagues had conducted a genetic screen in yeast to discover genes whose activity affected the toxicity of alpha-synuclein. That study showed that genes enhancing ER-to-Golgi trafficking prevented alpha-synuclein toxicity. In particular, they found that one protein, called Ypt1p, which is involved in regulating trafficking could also be switched on to suppress alpha-synuclein toxicity in yeast cells.

Our findings indicated that this ER-to-Golgi trafficking pathway is intimately coupled to the pathology, although in humans there are likely others involved as well, given how many genes we found that modified alpha-synuclein toxicity, said Lindquist. But these findings were so persuasive that we decided we needed to test whether enhancing Ypt1p activity would suppress alpha-synuclein toxicity in animal models of the disease. Fortunately, we had an excellent team of collaborators with expertise in these models, who could conduct these studies.

The researchers next studied whether enhancing activity of the mammalian Ytp1p counterpart, called Rab1, suppressed alpha-synuclein toxicity in the fruitfly *Drosophila*, the roundworm *C. elegans* and in cultures of rat neurons. Bonini and her colleagues tested the effect in fruitflies; co-author Guy Caldwell and his colleagues at the University of Alabama performed the tests in roundworms; and co-author Jean-Christophe Roche and his colleagues at Purdue performed the tests in rat neurons. Caldwell is also coordinator of HHMI's Undergraduate Research Intern Program at the University of Alabama.

They all came back with the same answer, said Lindquist. All saw significant suppression of toxicity; although none saw complete suppression, which confirms our yeast studies showing that other pathways are affected by alpha-synuclein accumulation. However, importantly, the results of our genetic screen have given us a way to ask important questions about these other aspects of alpha-synuclein toxicity, she said.

Lindquist also said the findings give important clues to why dopamine-producing neurons in the brain are the most vulnerable neurons to toxic alpha-synuclein accumulation. The death of such neurons reduces brain dopamine levels, causing the tremors and other symptoms of Parkinson's disease. Dopamine is one of many types of neurotransmitter — chemical

signals that one neuron launches at its neighbor to trigger a nerve impulse.

Of all the neurotransmitters, dopamine has a higher potential for being toxic, she said. Its toxicity is normally prevented in the neuron by sequestration within vesicles for transport from the ER. But a defect in ER trafficking caused by alpha-synuclein accumulation could cause the toxic buildup of dopamine to occur in these neurons.

Lindquist and her colleagues believe their findings will guide the search for new drugs that suppress alpha-synuclein toxicity by enhancing the machinery of ER-to-Golgi transport. Thus, she said, they have already conducted a screen of 150,000 compounds for those with therapeutic potential.

We have found compounds that reverse alpha-synuclein toxicity, and we plan to publish those results soon, she said. These findings are exciting because they tell us we have a platform for discovering new therapeutic strategies and for speeding the process of discovering treatments for these disorders.

Current treatments for Parkinson's disease do not aim at protecting the dopamine-producing neurons themselves. Rather, the treatments seek to restore dopamine levels in the brain or to treat symptoms of the disease.

Lindquist cautioned that the findings have not by any means proven that this mechanism of pathology or the compounds that affect it are relevant to humans. However, given the fact that we've found the same results in yeast, flies, worms and rat neurons, I would be very surprised if we didn't find that they were relevant in humans, she said.

Bonini added that the research team's findings illustrate the power of animal models in revealing insight into Parkinson's disease. These results highlight the value and importance of very simple model organisms in studying these disorders, she said. For example, yeast is only a single cell, not even a neuron, and yet it can reveal proteins that modify the toxicity of alpha-synuclein. And in flies, it is possible to study the effects of these proteins on alpha-synuclein toxicity in dopaminergic neurons. Clearly, these kinds of basic research collaborations, in which you can progress up the evolutionary tree using multiple model organisms, will open the door to new therapeutic opportunities for Parkinson's, said Bonini.