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Untangling a Link Between Normal Protein Folding and Alzheimer's Disease

An enzyme that snips apart proteins that form brain-clogging plaques in people with Alzheimer's disease also appears to regulate enzymes that fold new proteins into their working forms in healthy cells.

The discovery offers new hints about how mutations or exposure to chemicals that affect the regulation of protein-folding machinery might stimulate a protein-snipping enzyme, called presenilin-1, which has been implicated in the pathogenesis of Alzheimer's disease. Such a link, if further confirmed, could have important implications for understanding and treating Alzheimer's disease, say the researchers.

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- Peter Walter

The research team, which included Howard Hughes Medical Institute (HHMI) investigators [Peter Walter](#) of the University of California, San Francisco (UCSF) and Randall Kaufman of the University of Michigan, as well as UCSF colleagues Maho Niwa and Carmela Sidrauski, reported its findings in an article in the December 22, 1999, issue of the journal *Cell*.

The researchers began their studies in hopes of learning more about how proteins involved in the "unfolded protein response" (UPR) detect the amount

of unfolded proteins in a cell and signal genes to either increase or decrease the production of protein-folding enzymes. Such signals are critical because newly synthesized proteins, which are essentially linear strings of amino acids, are functionally useless unless they are folded into a three-dimensional form.

The researchers knew that the UPR hinges on a protein, called Ire1, that senses the amount of unfolded proteins and switches on protein-folding genes. They suspected that Ire1 works in the nucleus where it cuts a specific messenger RNA (mRNA) at two places, so that it can be restitched into a gene-activating form by another enzyme.

Since studies of Ire1 splicing had only been done in yeast, the scientists first wanted to see whether such splicing occurred in mammalian cells. Thus, in their initial experiments they inserted the yeast mRNA into human cells and found that it was cut and spliced just as in yeast cells.

"This is a first report showing that salient features of this highly unusual signaling pathway are conserved in mammalian cells," said Walter.

The scientists next wanted to learn how Ire1, which is normally nestled in the membrane of the endoplasmic reticulum (ER), extends its activity all the way to the cell's nucleus. The enzyme appears to extend a "sensor" into the protein-synthesizing region of the ER, where it detects unfolded proteins. The other end of Ire1 that bears the RNA-splicing machinery extends toward the cytosol.

The scientists theorized that Ire1's RNA-splicing end might actually be snipped off, and like an enzymatic guided missile, enter and penetrate deep into the nucleus to splice the target mRNA.

In experiments designed to track the location of the Ire1-cleaved fragment within mammalian cells, the scientists found that the fragment did, indeed, invade the nucleus, and not remain part of the membrane-bound protein.

"This was a complete surprise that when we induced UPR in these cells, a large fraction of Ire1 localizes to the nucleus," said Walter.

"However, it was not all or nothing," he emphasized. "Even uninduced cells showed some Ire1 in the nucleus, so there are many complexities of the process we don't understand." Walter said that the two similar forms of Ire1, dubbed alpha and beta, might be regulated differently and have slightly different functions.

The scientists next sought the identity of the enzyme that snipped off the "missile" portion of Ire1, in a protein-cleaving reaction called proteolysis. Beginning with a hunch that presenilin-1 might be that enzyme, they obtained engineered cells that lacked presenilin-1 from Dennis Selkoe's laboratory at Harvard Medical School. They then studied the cells to see what effect, if any, the loss of presenilin-1 activity would have on Ire1, and found that cells without presenilin-1 did not show proper movement of Ire1 into the nucleus.

"While this analysis establishes a link between presenilin-1 and Ire1 processing, it is not necessarily a direct one, since we have not figured out its biochemistry," emphasized Walter.

Presenilin-1's apparent role in protein folding links this normal cellular process to Alzheimer's disease because researchers had previously shown that presenilin-1 is part of the machinery that slices apart "amyloid precursor protein" to produce the amyloid plaques that clog the brains of Alzheimer's patients. Presenilin-1's dual role could aid in both understanding and treating Alzheimer's disease, said Walter.

For example, he said, abnormally activated Ire1 -- perhaps through genetic mutation -- could overstimulate presenilin-1, which could act to create amyloid plaque deposition.

"Also, environmental agents or toxins could cause protein misfolding through the UPR might induce presenilin-1 activity, which in turn might activate a proteolytic cascade that could also lead to increased amyloid deposits," said Walter.

"Finally, this indication that presenilin-1 plays a role in normal protein processing makes it unlikely that Alzheimer's disease could be treated using drugs to block this pathway without severe side effects for normal physiology," said Walter.