Researchers unveil near-complete protein catalog for mitochondria

July 10 2008

Imagine trying to figure out how your car's power train works from just a few of its myriad components: It would be nearly impossible. Scientists have long faced a similar challenge in understanding cells' tiny powerhouses — called "mitochondria" — from scant knowledge of their molecular parts.

Now, an international team of researchers has created the most comprehensive "parts list" to date for mitochondria, a compendium that includes nearly 1,100 proteins. By mining this critical resource, the researchers have already gained deep insights into the biological roles and evolutionary histories of several key proteins. In addition, this careful cataloging has identified a mutation in a novel protein-coding gene as the cause behind one devastating mitochondrial disease. A full description of the work appears in the July 11 print edition of the journal *Cell*.

"For years, a fundamental question in cell biology has gone largely unanswered — what proteins function in mitochondria?" said Vamsi Mootha, an associate member at the Broad Institute of Harvard and MIT and a Harvard Medical School assistant professor at Massachusetts General Hospital, who led the study. "By creating a comprehensive list, we now have a valuable resource that has already helped enhance our understanding of mitochondrial biology and disease."

Mitochondria are linchpins of cellular life, found within the cells of all eukaryotes from yeast to humans. These miniaturized organs

("organelles") are well known for their role in providing cellular energy. They have also been implicated in a wide range of normal and disease processes, including diabetes, neurodegeneration, cancer, drug toxicity and aging.

Although mitochondria have their own genome — a vestige from their days as free-living bacteria — the vast majority of the critical mitochondrial proteins are derived not from their genome, but rather from the nuclear genome. However, even with the wealth of genome sequence data now available, scientists have struggled to identify which genes encode the roughly 1,200 proteins that make up a functional mitochondrion.

Researchers from the Broad Institute, Harvard Medical School, and Massachusetts General Hospital worked together to address this problem, drawing on the power of a multi-faceted approach that includes large-scale, mass spectrometry-based proteomics to measure proteins in mitochondria from a variety of tissues; computational methods to help identify those proteins that cannot be reliably detected; and microscopy to confirm within human cells the localization of presumptive mitochondrial proteins.

"The technologies and analytical methods for measuring proteins on a large scale are really transforming what we can learn about human biology," said Steve Carr, director of the Proteomics Platform at the Broad Institute and a co-author of the Cell paper. "By applying them to mitochondria isolated from fourteen different mouse tissues, we've completed one of the most comprehensive proteomic analyses of any organelle to date."

As a result of their analyses, the researchers identified a total of 1,098 mitochondrial proteins to form a compendium they have named "MitoCarta," and which is available to the entire scientific community.

Notably, about one-third of this inventory has not been previously linked to the organelle.

To shed light on the functions of the newly uncovered mitochondrial proteins, the researchers compared the proteins' corresponding gene sequences across hundreds of species, from humans and fish to fungi and bacteria. "Proteins with similar roles often share similar histories, meaning they're gained or lost together during evolution," said Mootha. "We decided to use this tendency to our advantage to decipher how some mitochondrial proteins work."

By examining the organelle's proteins through this evolutionary lens, the researchers uncovered a striking pattern. A group of key mitochondrial proteins, known to be absent in yeast but otherwise present among eukaryotes, are actually missing from several other single-celled species. In organisms that have them, including humans and other mammals, these proteins contribute to a boot-shaped, multi-protein structure, which forms the gateway to a critical step in the energy-generation process. By virtue of these proteins' shared — and unusual — past, Mootha and his colleagues were able to identify several additional proteins that are also associated with this crucial mitochondrial structure, known as complex 1.

In addition to offering insights into mitochondrial biology, these discoveries also paved the way for a breakthrough in understanding mitochondrial disease. For decades, doctors have diagnosed patients with deficiencies in complex I function. These disorders affect about 1 in 5,000 newborns, are genetic in origin, and are lethal in the first few years of life. Yet for many cases a culprit gene cannot be found. However, thanks to MitoCarta and its corresponding evolutionary analyses, the researchers and their collaborators at the University of Melbourne and Royal Children's Hospital in Australia identified a mutation in a novel gene, called C8orf38, as one cause of complex I disease.

"Our finding underscores the power of this protein catalogue to open new vistas on disease," said Mootha. "It promises to shed light not only on rare metabolic diseases, but common diseases as well."

Source: Broad Institute of MIT and Harvard