TECHNIQUES

Many makes of mitochondria

> mitochondria in neuronal and glial cells might be specialized for different functions

Mitochondria have many important functions in the CNS, but do they show some functional diversity across different types of neural cells? Fecher et al. describe a method that enables mitochondria from specific cell types to be captured and compared. Using this method, they reveal differences in protein expression and function between mitochondria of different cerebellar cell types in the mouse brain.

The authors generated MitoTag mice, which show Cre-recombinase-dependent expression of green fluorescent protein (GFP) that is targeted to the outer membrane of mitochondria. Crossing these mice with mice expressing Cre under the control of a cell-type marker generated mice with cell-type-specific labelling of mitochondria. To capture cell-type-specific mitochondria from a tissue mixture of labelled and unlabelled mitochondria, the authors added magnetic microbeads that selectively bind to GFP, and applied a magnetic field to separate out the GFP-labelled organelles.

Credit: Jose A. Bernat Bacete/Getty

Using this immunocapture technique, the authors isolated mitochondria from three different cell types in the mouse cerebellum: Purkinje cells (inhibitory neurons), granule cells (excitatory neurons) and astrocytes. Once isolated, the authors screened the proteomes of these mitochondria for comparison.

More than 85% of the proteins identified were similarly abundant in all three cell types, suggesting that many mitochondrial functions are conserved between the cell types. However, almost 200 of the mitochondrial proteins identified were differentially regulated across the three groups. A comparison of the mitochondrial proteomes of the two neuronal types with that of the astrocytes using gene ontology databases indicated that mitochondria in neuronal and glial cells might be specialized for different functions. For example, whereas neuronal mitochondria are specialized for generating the metabolite ubiquinone, astrocytic mitochondria seem better equipped to metabolize lipids. Further experiments comparing the ability of mitochondria to metabolize long-chain fatty acids confirmed that astrocytic mitochondria are indeed more efficient at metabolizing such molecules than are Purkinje-cell-derived mitochondria.

The authors also found that granule-cell-derived mitochondria expressed higher levels of the mitochondrial calcium uniporter MCU than Purkinje-cell-derived or astrocyte-derived mitochondria. Consistent with this finding, isolated mitochondria from granule cells were able to buffer calcium more efficiently than mitochondria from the other two cell types.

Purkinje-cell mitochondria were enriched for a protein that establishes connections between the endoplasmic reticulum (ER) and mitochondria. Confirming the functional relevance of this, the cell bodies of Purkinje cells showed more ER–mitochondrial contacts than did astrocytes or granule cell bodies. Thus, proteomic differences identified using this isolation technique can predict differences in mitochondrial function, even between neuronal cell types.

One potential use for this technique might be to examine disease-related changes in cell-type-specific mitochondria. Fecher et al. labelled markers of neuronal and astrocytic mitochondria (identified through their proteomic analyses) in tissue samples from mouse models of disease. In samples of cortex from APP/PS1 mice, a model of Alzheimer disease, the number of neuronal (and, to lesser extent, glial) mitochondria around amyloid-β plaques seemed to be reduced. Moreover, in spinal cords of pre-symptomatic *SOD*^{G93A} mice — a model of amyotrophic lateral sclerosis (ALS) — neuronal, but not glial, mitochondria showed morphological changes compared with those in wild-type mice. Notably, these mitochondrial changes were confirmed by labelling cell-type-specific mitochondria in post-mortem samples from individuals with Alzheimer disease or ALS, suggesting possible translational relevance of this approach.

Together, the data obtained using the authors' MitoTag and immunocapture method have revealed mitochondrial diversity in the CNS. Further use of this technique might lead to a greater understanding of why some cell types are more vulnerable than others to neurological diseases associated with changes in mitochondrial function.

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