Quantitative analysis of the mitochondrial proteome and phosphoproteome in the yeast *Saccharomyces cerevisiae*

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The mitochondrion is an organelle of which the most important function is to provide energy to the cell generated by oxidative phosphorylation catalyzed by the respiratory enzymes. In humans, deregulation of mitochondrial functions is associated with several pathologies. The activity of the respiratory enzymes may be modulated in response to metabolic demand and various types of stress. Several levels of regulation may be conceived, including post-translational modifications, such as phosphorylation. The steadily increasing number of identified mitochondrial phosphoproteins suggests that reversible protein phosphorylation could be an important level of regulation in mitochondria. However, this hypothesis cannot be tested without quantitative data on variations in the abundance of mitochondrial proteins and their level of phosphorylation under different growth conditions. The yeast *Saccharomyces cerevisiae* is a powerful tool for studying various energetic and physiological states. We present for the first time a quantitative study of both protein abundance and phosphorylation levels in isolated yeast mitochondria under respiratory and fermentative conditions. To focus our analysis specifically on mitochondrial proteins, we performed a subcellular fractionation and used LC–MS/MS to overcome the limitations of 2D gel electrophoresis. Protein abundances were quantified using a label-free method. The phosphoproteome was analyzed quantitatively using the multiplex stable isotope dimethyl labeling procedure. For all quantified phosphopeptides, protein abundance was determined, allowing normalization of the data and permitting analysis of the specific variation of phosphorylation status independent of changes in protein abundance. This study provided reliable information on how the yeast mitochondrial proteome and phosphoproteome adapt to different carbon sources.

Biography

Lemaire Claire is a biochemist specialized in membrane proteins. She began her career in the photosynthesis field on the assembly and regulation of photosynthetic complexes (I.B.P.C., Paris). She then joined the C.N.R.S. (French National Center for Scientific Research) where she acquired a solid expertise in the respiratory complexes. Over the last six years, she has developed a research project with her group focusing on the regulation of OXPHOS complexes by phosphorylation.

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