The PINK1 kinase-driven ubiquitin ligase Parkin promotes mitochondrial protein import through the presequence pathway in living cells

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Résumé

Parkinson's disease (PD) is characterized by the specific degeneration of the dopaminergic neurons of the *substantia nigra*. Autosomal recessive PD forms are caused by mutations in the genes encoding the E3 ubiquitin ligase Parkin (*PARK2*) and the mitochondrial serine/threonine-protein kinase PINK1 (*PARK6*). These proteins cooperate in the regulation of various mitochondrial quality pathways. Our team has contributed to demonstrate that the PINK1/Parkin system couples the mitochondrial protein import process through the TOM machinery (translocase of outer mitochondrial membrane) to mitophagy. Mitochondrial import efficiency reflects the degree of mitochondrial dysfunction, thereby determining the activation of the PINK1/Parkin system. Mitochondrial import has more generally emerged as a key element of the mitochondria to nucleus communication in response to mitochondrial stress. We engineered an inducible biosensor for monitoring the main presequence-mediated import pathway with a quantitative, luminescence-based readout. We used this tool to explore the regulation of mitochondrial import by the PINK1 kinase-driven Parkin ubiquitin ligase. We found that mitochondrial import was stimulated by Parkin, but not by disease-causing Parkin variants. This effect was dependent on Parkin activation by PINK1 and accompanied by an increase in the abundance of ubiquitin chains on mitochondria and by ubiquitylation of subunits of the TOM complex. Mitochondrial import efficiency was abnormally low in cells from patients with PINK1-and PARK2-linked Parkinson's disease and was restored by phosphomimetic ubiquitin in cells with residual Parkin activity. Altogether, these findings uncover a role of ubiquitylation in mitochondrial import regulation and suggest that loss of this regulatory loop may underlie the pathophysiology of Parkinson's disease.

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