NEURODEGENERATIVE DISEASE

Pink, parkin and the brain

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Dysfunctions in a number of cellular pathways can cause Parkinson's disease. Fruitflies with mutations in a protein called PINK1 show that there might be some unsuspected interplay between two such pathways.

Parkinson's disease was first described[1] in 1817, but our understanding of what causes the neurodegeneration that underlies its devastating symptoms is still rudimentary. Such poor understanding hinders the development of therapies, which currently don't seem to modify disease progression, even if they can mitigate for a time some of the movement difficulties that characterize this condition. In this issue, Clark *et al.* (page 1162)[2] and Park *et al.* (page 1157)[3] provide clues to the basis of degeneration in Parkinson's disease by linking two causative mechanisms that were previously thought to be separate.

Most cases of typical 'sporadic' Parkinson's disease are believed to result from a lifetime of environmental exposures superimposed on an individual's genetic susceptibilities. However, a small fraction of cases (probably less than 10%) is caused by single-gene mutations. These provide insights into the cellular pathways involved in neurodegeneration[4]. For example, the products of two of the genes mutated in Parkinson's disease — parkin and ubiquitin carboxy-terminal hydrolase L1 — are components of the ubiquitin–proteasome system (UPS) that degrades damaged or misfolded proteins. Moreover, duplication or triplication of the normal alpha-synuclein gene causes Parkinson's disease, and overexpression of alpha-synuclein inhibits UPS function. Together, this evidence strongly implicates dysfunction of UPS in the development of Parkinson's disease.

Other single-gene mutations point to Parkinson's disease being associated with defects in mitochondria — the organelles that carry out respiration in the cell. Chemicals that inhibit the mitochondrial 'complex I' can reproduce many features of Parkinson's disease in experimental systems[5]. The mitochondrial hypothesis of Parkinson's disease was put on even firmer footing with the discoveries of causative mutations in the *DJ-1* and *PINK1* genes. The DJ-1 protein has a role in the oxidative stress response; under oxidative conditions, some of the cellular DJ-1 moves to mitochondria where its function remains to be elucidated. The PINK1 protein is found primarily in mitochondria, and it is predicted to be a kinase (an enzyme that adds phosphate groups to other proteins), although its substrates are unknown[6].

So, it looked as though there was the beginning of a nice, neat parcelling of two distinct mechanisms that cause Parkinson's disease: UPS dysfunction and mitochondrial impairment. Of course, things are rarely so simple, and the first inkling that things were not quite as they seemed came from studies showing that overexpression of parkin in cultured cells delays toxin-induced mitochondrial dysfunction[7] and that fruitflies that lack the *parkin* gene have prominent mitochondrial abnormalities in many tissues[8]. Mitochondrial defects were also observed in parkin-deficient mice and humans[9,10]. Why would loss of this protein cause mitochondrial dysfunction?

Now Clark *et al.*[2] and Park *et al.*[3] raise additional questions about what parkin is — or isn't — doing to cause Parkinson's disease, although the primary focus of these papers is the *PINK1* gene. Both papers show that fruitflies bearing mutations in the fly version of *PINK1* display degeneration of flight muscles and defective sperm formation. Using a combination of biochemical and imaging approaches, these researchers further report that mitochondrial defects accompany both of these abnormalities. Park *et al.*[3] also report that the *PINK1*-mutant flies show loss of dopamine neurons — a type of neuron known to degenerate in Parkinson's disease — with an accompanying mitochondrial swelling. Although the specific tissues affected by loss of PINK1 function differ in flies and humans, the fact that each of the tissues affected by *PINK1* mutations in the fruitfly has mitochondrial defects strongly suggests that the neurodegeneration in humans with *PINK1* mutations is also caused by mitochondrial dysfunction. This finding is interesting, but not unexpected.

The real surprise and importance of these papers is the finding that parkin seems to act downstream from PINK1 in a common pathway that influences mitochondrial integrity. Indeed, the strikingly similar characteristics of the *PINK1*- and *parkin*-mutant fruitflies, including flight-muscle degeneration, sperm-formation defects and mitochondrial abnormalities, alone argue that these two genes act in a common pathway. Moreover, both papers show that overexpression of parkin compensates for a lack of PINK1, preventing the effects of the *PINK1* mutation. However, PINK1 overexpression does not detectably influence the characteristics of the *parkin* mutants. Both papers also show that *PINK1–parkin* double mutants have symptoms that are indistinguishable from those seen in the single mutants. By contrast, the *DJ-1*-mutant fruitflies are quite different from *parkin* and *PINK1* mutants, suggesting that DJ-1 influences different pathways. Interestingly, the similarities between the *PINK1*- and *parkin*-mutant fruitflies parallel a recent clinical study showing that in humans *PINK1* and *parkin* mutations can produce similar symptoms[11].

What is the nature of the pathway regulated by PINK1 and parkin? The simplest interpretation of the data is that PINK1 decreases parkin abundance by reducing the level of parkin messenger RNA or protein. Alternatively, PINK1 may phosphorylate parkin and directly influence its activity. However, a problem with the latter model is that most data suggest that PINK1 protein resides primarily in mitochondria and that parkin lies outside mitochondria. Nevertheless, it remains possible that the localization of either parkin or PINK1 changes upon stress, and there are several reports, including that from Park *et al.*[3], that have argued that at least some parkin associates with mitochondria.

Another model to explain the current findings is that PINK1-induced mitochondrial impairment leads to secondary dysfunction of the UPS, which can be ameliorated by overexpression of parkin. Parkin, a component of the UPS, is thought to act as a ubiquitin E3 ligase, an enzyme that directs proteins destined for destruction to the proteasome by tagging them with a ubiquitin group. In this regard, there is evidence that parkin mutations are associated with complex I defects[10] and that complex I defects can inhibit the UPS in animal models of Parkinson's disease[12].

Finally, it remains possible that parkin has a functional role in mitochondria that does not involve its ubiquitin-ligase activity. Although there is substantial evidence *in vitro* that parkin can work as a ubiquitin ligase, few of the reported substrates of parkin have been validated *in vivo*. Furthermore, there is evidence that parkin can regulate the biogenesis of mitochondria[13]. Future experiments to delineate the PINK1–parkin pathway should clarify the mechanisms underlying neurodegeneration in Parkinson's disease and shed light on some very basic questions of mitochondrial biology. ■

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