

Metabolism and the Other Fat: A Protocadherin in Mitochondria

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The protocadherin Fat is known as a tumor suppressor regulating growth in *Drosophila* and for its conserved function during planar cell polarity establishment. McNeill and colleagues now identify an unsuspected role for a C-terminal proteolytic product of Fat in mitochondria: regulating the electron transport machinery and metabolism.

In a surprising new paper in this issue of *Cell*, Sing et al. (2014) report that the mitochondrial electron transport chain depends directly on a polypeptide imported into the mitochondrion after cleavage from a large transmembrane receptor at the cell surface, Fat (Ft). This Ft^{Mito} species helps to assemble or maintain complexes I and V, required for oxidative phosphorylation and ATP generation. This is all the more remarkable because Ft was hardly an unstudied protein but for some years has been the focus of intense attention for its roles in growth control, tumor suppression, and planar cell polarity (PCP) (Figure 1; Thomas and Strutt, 2012; Sharma and McNeill, 2013). This provocative new study suggests that these processes may be coupled to mitochondrial function and signaling and raises numerous questions about the function and regulation of mitochondria.

The protocadherin Ft together with the ligand Dachshous (Ds) forms one of the two systems that establish PCP, the asymmetry of cells within the plane of the epithelium, and thus controls the formation of asymmetric structures and morphogenetic cell behaviors, a function likely conserved between *Drosophila* and vertebrates (Figure 1; Sharma and McNeill, 2013). Ft also regulates growth through the conserved Salvador-Warts-Hippo (SWH) pathway of tumor suppressors by inhibiting the transcription factor York1 (the homolog of mammalian Yap and Taz). Neither the molecular mechanism connecting Ft to the SWH pathway nor its conservation are yet fully elucidated (Bossuyt et al., 2014).

Sing et al. started by characterizing potential Ft-binding proteins recovered from a two-hybrid screen, using RNAi experiments in vivo. Surprisingly, three interactors showing PCP phenotypes were mitochondrial proteins: NADH dehydrogenase ubiquinone flavoprotein 2 (Ndufv2), mitochondrial processing protease (Mpp), and the V-ATPase component CG1746. Subsequent studies showed that, like *ft* mutants, Ndufv2 knockdown impairs SWH signaling. Remarkably, a fraction of both transfected and endogenous Ft colocalized with the mitochondrial marker CVa and Ndufv2 in cultured cells. Cell fractionation revealed that mitochondrial Ft corresponds to a 68 kDa cleavage product (Ft^{Mito}), derived from the Ft intracellular domain and residing inside of mitochondria. Ft^{Mito} may be produced by a cleavage occurring at the cell surface, as it is not produced unless the Ft intracellular domain is expressed with its transmembrane segment.

Sing et al. then made use of the characteristic large mitochondria found in developing sperm and saw that mitochondria from *ft* mutant cells are swollen and rounded and lack proper cristae morphology. These cell-autonomous structural defects are accompanied by functional deficiencies assayed in imaginal disc tissues, where superoxide levels and markers of oxidative stress were elevated by mutation of *ft*, as were cellular lactate levels, indicating a higher level of glycolysis accompanying the reduction in mitochondrial complex I activity.

The *ft* mutant phenotype is comparable to that caused by loss of a known complex I assembly factor. Intriguingly, Ft^{Mito} is detected in intact complex I. Although levels of individual complex I proteins are not affected in *ft* mutants, levels of the intact complex are reduced significantly. Levels of complex V are diminished also, although Ft^{Mito} itself is not detected in complex V.

The authors hypothesize that Ft connects growth with metabolism: cleavage of the Ft^{Mito} fragment, which may prevent Ft from inhibiting growth through the SWH pathway, enhances assembly of complex I and complex V, thereby enhancing oxidative phosphorylation at the expense of glycolysis. What would be the purpose of such a shift? Although ATP might be required to support the growth that should accompany reduced SWH signaling, the authors did not see much effect of *ft* mutations on ATP levels, which may be partially compensated by increased glycolysis. In addition, it is generally thought that glycolysis rather than oxidative phosphorylation is the favored energy source for growing cells, as glycolysis, accompanied by citrate export from mitochondria, provides building blocks for biosynthesis of nucleotides, amino acids, and lipids, in contrast to oxidative phosphorylation, which burns carbon through the TCA cycle (Vander Heiden et al., 2009). The authors expect, therefore, that processing of Ft for mitochondrial entry may mitigate the enhanced growth signals expected as a result of diminished tumor suppression with a switch away from

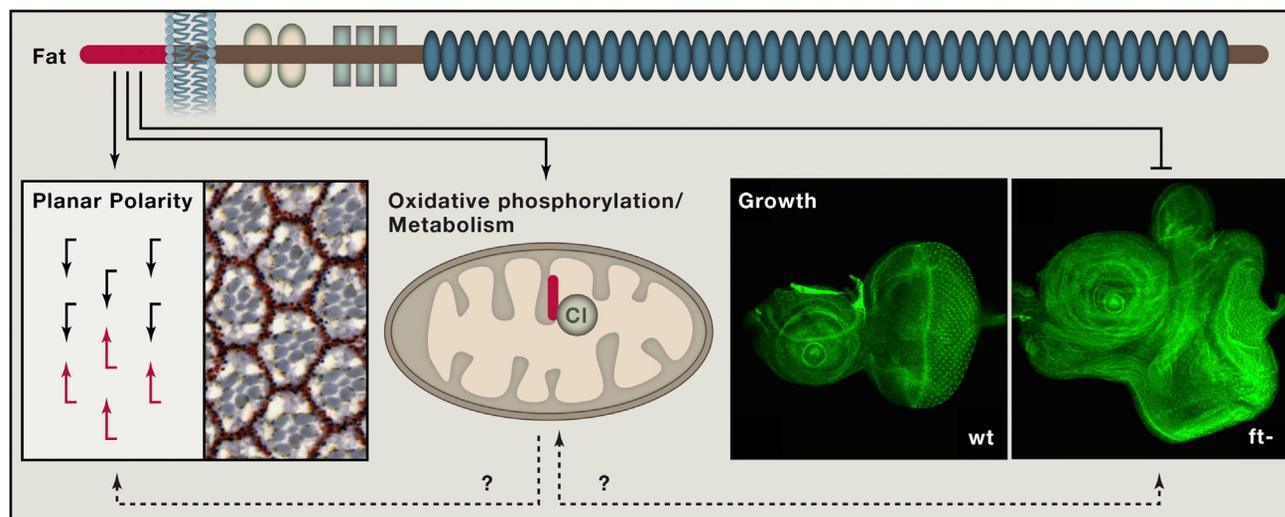


Figure 1. The Diverse Functions of Fat

The protocadherin Fat (top) has long been known to regulate the establishment of planar cell polarity. For example, the planar polarity of cells in the *Drosophila* eye is reflected in the asymmetry of ommatidial clusters; the distribution of two chiral forms is summarized on the left. Fat also acts as tumor suppressor repressing growth (the wild-type eye-antennal imaginal disc (right) is smaller than the *ft* mutant). Sing et al. now have identified a novel role for a C-terminal proteolytic fragment of Fat in oxidative phosphorylation in mitochondria (center). The possible interplay between the different functions awaits further elucidation, as discussed in the text. Eye disk images have been adapted from Zhao et al. (2013).

glycolysis toward oxidative phosphorylation. This is to be contrasted with the effects of *ft* mutations, wherein growth promotion would be accompanied by enhanced dependence on glycolysis. Many human tumors have mutations in Fat homologs (Morris et al., 2013).

These findings raise a multitude of questions. What, for example, is the stoichiometry of Ft processing? Is sufficient Ft^{Mito} produced to significantly affect the capacity for SWH and PCP signaling? Is Ft^{Mito} a constituent of the complex I, and does every complex I contain Ft polypeptides? Because complex I assembly is not completely lost in *ft* mutants and Ft has not been identified in proteomic studies of mitochondria (Pagliarini et al., 2008), it is also possible that Ft^{Mito} is a regulator or assembly factor. Does the location of full-length Ft on the cell surface and its believed role as a cell surface receptor in PCP and in growth control imply that mitochondria are regulated by extracellular cues through Ft? Curiously, mutants for Ft's extracellular ligand Dachshous do not show a parallel reduction in complex I assembly, leaving this question open. Last but not least, what regulates mitochondria in nonepithelial tissues or in organisms that lack Ft proteins, such as *C. elegans* or *S. cerevisiae*?

Do these roles of Ft imply functional connections between mitochondrial function, SWH tumor suppressor signaling, and PCP (Figure 1)? The molecular mechanism connecting Ft to the SWH pathway has been far from clear, raising the possibility that Ft signaling to SWH is indirect and is mediated by mitochondria. It is intriguing that knockdown of other mitochondrial components leads to PCP defects and changes in SWH activity, which could indicate roles for mitochondria in PCP and growth control, and that there appears to be a gradient of ROS levels within the developing eye that resembles the gradient of Ft activity responsible for PCP. For the time being, however, Sing et al. take the conservative view that mitochondrial stability, PCP, and tumor suppression may be largely separable, chiefly because conserved Ndufv2-binding sequences within the Ft intracellular domain are required for its mitochondrial function, but not for growth suppression through the SWH pathway and for PCP. In their view, some of the unexplained effects of mitochondria on PCP and SWH signaling may reflect feedback pathways. These data, like previous structure-function analyses of Ft, are not clear-cut, however, so the last chapter may not have been written on this interesting topic.

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REFERENCES

- Bossuyt, W., Chen, C.L., Chen, Q., Sudol, M., McNeill, H., Pan, D., Kopp, A., and Halder, G. (2014). *Oncogene* 33, 1218–1228.
- Morris, L.G., Ramaswami, D., and Chan, T.A. (2013). *Cell Cycle* 12, 1011–1012.
- Pagliarini, D.J., Calvo, S.E., Chang, B., Sheth, S.A., Vafai, S.B., Ong, S.E., Walford, G.A., Sugiana, C., Boneh, A., Chen, W.K., et al. (2008). *Cell* 134, 112–123.
- Sharma, P., and McNeill, H. (2013). *Prog. Mol. Biol. Transl. Sci.* 116, 215–235.
- Sing, A., Tsatskis, Y., Fabian, L., Hester, I., Rosenfeld, R., Serricchio, M., Yau, N., Bietenhader, M., Shanbhag, R., Jurisicova, A., et al. (2014). *Cell* 158, this issue, 1293–1308.
- Thomas, C., and Strutt, D. (2012). *Dev. Dyn.* 241, 27–39.
- Vander Heiden, M.G., Cantley, L.C., and Thompson, C.B. (2009). *Science* 324, 1029–1033.
- Zhao, X., Yang, C.H., and Simon, M.A. (2013). *PLoS One* 8. Published online May 7 2013. <http://dx.doi.org/10.1371/journal.pone.0062998>.