Fussy mitochondria fuse in response to stress

Alexander M van der Bliek

Department of Biological Chemistry, David Geffen School of Medicine at University of California Los Angeles, Los Angeles, CA, USA. Correspondence to: avan@mednet.ucla.edu

The EMBO Journal (2009) 28, 1533–1534. doi:10.1038/emboj.2009.130

When cells become committed to apoptosis, they shatter their mitochondrial networks through the actions of the mitochondrial fission protein DRP1. Massive fragmentation of mitochondria facilitates simultaneous release of cytochrome *c* from all mitochondria within a cell, thus promoting further progression along the apoptotic pathway. In this issue, Tondera *et al* (2009) describe a new process with the opposite effect. When cells are subjected to modest levels of stress (well below levels needed to induce apoptosis), their mitochondria fuse to each other forming a closed network, similar to networks observed when mitochondrial fission is blocked. Stress-induced mitochondrial hyperfusion (SIMH), as this process was called, might counter stress by optimizing mitochondrial ATP production.

Mitochondria of healthy cells continually divide and fuse with each other (Okamoto and Shaw, 2005; Chan, 2006; Hoppins *et al*, 2007). Mitochondrial fission facilitates the redistribution of mitochondria in response to local changes in the demand for ATP and it allows for disposal of faulty mitochondrial fragments through mitophagy, whereas mitochondrial fusion promotes exchange of mtDNA and other vital components, thus reinvigorating the mitochondrial network. Mitochondrial fission and fusion processes are mediated by a series of dynamin family members. Fission is mediated by the dynamin-related protein DRP1. Mutations in the gene coding for this protein give rise to a highly interconnected network of mitochondria. Fusion is mediated by the dynamin-related proteins MFN1 and MFN2 at the mitochondrial outer membranes and the dynamin-related protein OPA1 at the mitochondrial inner membranes. Mutations in the genes coding for these proteins give rise to fragmented mitochondria, because fission still occurs while fusion is blocked. Mutations in the genes coding for the mitochondrial fusion proteins are also responsible for two human diseases. Patients with a heterozygous mutation in MFN2 develop a peripheral neuropathy called Charcot–Marie–Tooth disease (CMT-2A) and patients with heterozygous mutations in OPA1 develop dominant optic atrophy (DOA) through progressive loss of retinal ganglion cells. The severity of heterozygous mutations in mice show that mitochondrial fission and fusion processes are crucially important for cell survival (Chan, 2006; Davies *et al*, 2007).

The rates of mitochondrial fission and fusion vary between cell types and different growth conditions, but they are usually balanced within a cell. However, the rate of fission does increase markedly without a compensating increase in the

rate of fusion when cells become committed to apoptosis (Suen *et al*, 2008). Increased fission in apoptotic cells coincides with the release of cytochrome *c* and mutations in the genes for the fission protein DRP1 can delay the release of cytochrome *c*, suggesting that apoptotic cytochrome *c* release is intimately connected with mitochondrial fission. Tondera *et al* (2009) now add a new twist to this plot with their discovery of a pathway that they call Stress-induced mitochondrial hyperfusion (SIMH) pathway. They show that treatments with low levels of toxic agents such as cycloheximide, UV irradiation or actinomycin D have the opposite effect of full-blown apoptosis-inducing treatments. Instead of inducing mitochondrial fragmentation as observed in apoptotic cells, these treatments cause mitochondria to fuse into a closed network, similar to networks observed in cells with mitochondrial fission defects (Figure 1). This closed network confers some degree of

Figure 1 Mitochondrial fission and fusion cycles normally consist of fission mediated by DRP1 and fusion mediated by MFN1, MFN2 and the short and long forms (S and L) of OPA1. Cells that are committed to apoptosis (equated here with high levels of stress) use DRP1-mediated fission to promote cytochrome *c* release. The newly discovered SIMH pathway, which is induced by low levels of stress, has the opposite effect, namely the formation of a closed mitochondrial network through the actions of MFN1 and L-OPA1, but not MFN2 or S-OPA1. SIMH pathway confers resistance to further stress, but eventually cells might succumb to apoptosis.

resistance to further insults, perhaps by increasing the robustness of mitochondria through exchange of crucial components.

One might ask whether the SIMH pathway merely reflects increased activity of the conventional fusion machinery or alternatively represents a deliberate switch to an alternative pathway. Tondera *et al* (2009) found three differences in protein requirements, suggesting mechanistic differences between SIMH pathway and conventional fusion. The first difference was observed with *MFN1* and *MFN2* knockout cells, which normally have fragmented mitochondria. Fragmentation in both cell types indicates that conventional mitochondrial fusion depends on the combined actions of *MFN1* and *MFN2*. However, low levels of stress induce filamentous mitochondria in *MFN2* knockout cells but not in *MFN1* knockout cells, suggesting that SIMH pathway requires *MFN1* but not *MFN2* (Figure 1). Several years ago a specific requirement for *MFN1* was noted when fusion was induced by the overexpression of *OPA1* (Cipolat *et al*, 2004). Could this inducible fusion have been equivalent to SIMH? Conversely, *MFN2* has not been linked with inducible fusion under either condition, but it was previously linked to Charcot-Marie-Tooth disease. The picture that is starting to emerge is that of dual requirements for *MFN1* and *MFN2* under normal conditions and additional dedicated functions of these proteins under special circumstances. *MFN2* could, for example, affect axonal transport of mitochondria, as suggested in the Charcot-Marie-Tooth disease, although *MFN1* can mediate mitochondrial fusion without help from *MFN2* when cells are subjected to low levels of stress.

The second difference was discovered with *OPA1*. In unstressed cells, certain isoforms of *OPA1* are constitutively cleaved by the mitochondrial intermembrane space AAA protease *YME1L* (Griparic *et al*, 2007; Song *et al*, 2007). This cleavage removes the amino terminal transmembrane segment to generate a short form of *OPA1* (S-*OPA1*). Isoforms that are not cleaved by *YME1L* retain their amino terminal transmembrane segment and are called the long form of *OPA1* (L-*OPA1*).

Conventional fusion of mitochondrial inner membranes requires the combined actions of S- and L-*OPA1* (Song *et al*, 2007). Tondera *et al* (2009) now show that L-*OPA1*, but not S-*OPA1*, is required for SIMH. A unique requirement for L-*OPA1* is consistent with an earlier report in which it was shown that *YME1L* siRNA, which leads to an accumulation of L-*OPA1*, also leads to a fused mitochondrial network (Griparic *et al*, 2007). The third difference was noted in the requirements for maintaining intact *OPA1*. Under adverse conditions, *OPA1* is proteolytically inactivated by the actions of an as yet unknown protease (Griparic *et al*, 2007). This proteolytic inactivation occurs during apoptosis, but it also occurs when certain scaffolding proteins called prohibitins are not around (Merkwirth *et al*, 2008). Tondera *et al* (2009) now show that SIMH requires a different scaffolding protein, the stomatin-like protein SLP-2, to prevent proteolytic inactivation of *OPA1*. As the requirements for *MFN1* and *MFN2* at the mitochondrial outer membrane, the requirements for S- and L-*OPA1* at the mitochondrial inner membrane and the scaffolding proteins required for maintaining the integrity of L-*OPA1* are all different, one can conclude that there are important differences between SIMH pathway and conventional mitochondrial fusion, consistent with an alternative pathway.

Until now, mitochondrial fusion seemed like a monolithic problem begging for a biochemical solution. The discoveries of Tondera *et al* (2009) add nuance to this problem. What are the specialized functions of *MFN1* and *MFN2*? Why does conventional fusion require S- and L-*OPA1*, whereas SIMH only needs L-*OPA1*? Why are different scaffolding proteins involved in *OPA1* function? In addition to these mechanistic questions, there are also new questions about regulation. How do different forms of cellular stress converge on the mitochondrial fusion apparatus? Which protein modifications steer the fusion apparatus towards the conventional fusion process and which ones steer it towards SIMH? These types of questions will inevitably change the course of future experiments.

References

- Chan DC (2006) Mitochondrial fusion and fission in mammals. *Annu Rev Cell Dev Biol* **22**: 79–99
- Cipolat S, Martins de Brito O, Dal Zilio B, Scorrano L (2004) *OPA1* requires mitofusin 1 to promote mitochondrial fusion. *Proc Natl Acad Sci USA* **101**: 15927–15932
- Davies VJ, Hollins AJ, Piechota MJ, Yip W, Davies JR, White KE, Nicols PP, Boulton ME, Votruba M (2007) *Opa1* deficiency in a mouse model of autosomal dominant optic atrophy impairs mitochondrial morphology, optic nerve structure and visual function. *Hum Mol Genet* **16**: 1307–1318
- Griparic L, Kanazawa T, van der Bliek AM (2007) Regulation of the mitochondrial dynamin-like protein *Opa1* by proteolytic cleavage. *J Cell Biol* **178**: 757–764
- Hoppins S, Lackner L, Nunnari J (2007) The machines that divide and fuse mitochondria. *Annu Rev Biochem* **76**: 751–780
- Merkwirth C, Dargazanli S, Tatsuta T, Geimer S, Lower B, Wunderlich FT, von Kleist-Retzow JC, Waisman A, Westermann B, Langer T (2008) Prohibitins control cell proliferation and apoptosis by regulating *OPA1*-dependent cristae morphogenesis in mitochondria. *Genes Dev* **22**: 476–488
- Okamoto K, Shaw JM (2005) Mitochondrial morphology and dynamics in yeast and multicellular eukaryotes. *Annu Rev Genet* **39**: 503–536
- Song Z, Chen H, Fiket M, Alexander C, Chan DC (2007) *OPA1* processing controls mitochondrial fusion and is regulated by mRNA splicing, membrane potential, and *Yme1L*. *J Cell Biol* **178**: 749–755
- Suen DF, Norris KL, Youle RJ (2008) Mitochondrial dynamics and apoptosis. *Genes Dev* **22**: 1577–1590
- Tondera D, Grandemange S, Jourdain A, Karbowski M, Mattenberger Y, Herzig S, Da Cruz S, Clerc P, Raschke I, Merkwirth C, Ehses S, Krause F, Chan DC, Alexander C, Bauer C, Youle R, Langer T, Martinou JC (2009) SLP-2 is required for stress-induced mitochondrial hyperfusion. *EMBO J* **28**: 1589–1600

This is an open-access article distributed under the terms of the Creative Commons Attribution License, which permits distribution, and reproduction in any medium, provided the original author and source are credited. This license does not permit commercial exploitation or the creation of derivative works without specific permission.

The EMBO Journal is published by Nature Publishing Group on behalf of European Molecular Biology Organization. This article is licensed under a Creative Commons Attribution-NonCommercial-No Derivative Works 3.0 Licence. [<http://creativecommons.org/licenses/by-nc-nd/3.0/>]