Cell Research (2010) 20:863-865.

www.nature.com/cr

RESEARCH HIGHLIGHT

A BID on mitochondria with MTCH2

Sara Cogliati[1], Luca Scorrano[1, 2]

[1]*Dulbecco-Telethon Institute, Venetian Institute of Molecular Medicine, Via Orus 2, 35129 Padova, Italy;* [2]*Department of Cell Physiology and Metabolism, University of Geneva, 1 Rue M. Servet, 1206 Geneve, Switzerland*
Cell Research (2010) **20**: 863-865. doi:10.1038/cr.2010.100; published online 13 July 2010

Apoptosis is a key process for tissue homeostasis and renewal. Its dysregulation is implicated in most human diseases, from cancer to neurodegeneration. Apoptosis is triggered by stimuli that damage the internal structures of the cell, or by specialized "death" receptors on its surface. In certain cell types, Bid, a "BH3-only" member of the Bcl-2 family of death regulators integrates these two pathways at the mitochondrial level. Despite years of intense research, the mechanisms by which Bid translocates to mitochondria remain unclear. A recent study by Gross and colleagues sheds new light on this process [1]. They identified MTCH2 as a mitochondrial protein that interacts with Bid and whose ablation dramatically affects mitochondrial translocation of this BH3-only protein. Interestingly, MTCH2 shares homology with members of the mitochondrial carrier family, but it is located on the outer membrane of the organelle; and it was recently reported to be associated with increased body mass index. Thus, this study not only unveils how BID is targeted to mitochondria during apoptosis, but also opens interesting avenues to investigate the relationship between mitochondria, apoptosis and control of metabolism.

Programmed cell death or apoptosis is a conserved pathway in all metazoans. It is fundamental in embryonic development, organogenesis and in maintaining tissue homeostasis in adult organisms. Impairment of apoptotic pathways leads to cancer, while their upregulation results in degenerative disease. In mammalian cells, there are two main pathways downstream of death signals that are linked in certain cell types: the "death receptor" pathway triggered by extrinsic stimuli (e.g. Fas, TNFα) and the mitochondrial pathway triggered by intrinsic death stimuli (e.g. DNA damage). Both culminate in the activation of caspases, cysteine proteases that cleave a number of substrates involved in maintenance of cytoskeletal and nuclear integrity, cell cycle progression and DNA repair, resulting in the orderly demise of the cell. Mitochondria participate in the competent activation of caspases, by releasing cytochrome *c* and additional apoptogenic factors from the intermembrane space into the cytosol. Cytochrome *c* in complex with Apaf-1 activates caspase 9 and other downstream "effector" caspases. The key regulators of this apoptotic process are proteins of the Bcl-2 family which orchestrate the signals leading to the activation of effector caspases [2]. In response to the activation of death receptors, the apical pro-caspase 8 undergoes autoproteolytic activation. In type I cells, such as thymocytes, active caspase 8 directly cleaves the effector caspases 3 and 7, whereas in type II cells, such as hepatocytes, the activation of the effector caspases requires the mitochondrial amplification loop. To this end pro-caspase 8 is recruited on mitochondria where it binds to cardiolipin [3]. There, after self activation induced by proximity, it cleaves the proapoptotic BH3-only member, BID, and activate it. The active form, christened truncated BID (tBID) on the surface of mitochondria, triggers the release of cytochrome *c* by causing oligomerization of BAX and BAK that results in outer membrane permeabilization [2] and by inducing Opa1-dependent cristae remodelling [4-6]. The importance of tBID in the Fas death pathway is well established, however the molecular mechanism of tBID recruitment on mitochondria is still unknown and this has been an area of intense investigation in the last years. Two main models have been put forward to explain the affinity of BID for mitochondria: one postulates that BID travels to mitochondria as a consequence of its affinity for specific lipids, or of its specific lipidation; the other involves the existence of one or more specific receptors on the mitochondrial surface that interact with tBID to assist its insertion in the mitochondrial membrane.

The first model is supported by earlier studies that indicated how N-terminal myristoylation increases the affinity of tBID for the organelle [7]. In addition, tBID binds to the phospholipid cardiolipin (retrieved only in mitochondria), which proved to be required for tBID

Correspondence: Luca Scorrano
E-mail: luca.scorrano@unige.ch

action [8]. Interestingly, cardiolipin on the outer mitochondrial membrane can also function as a scaffold for caspase 8, which translocates to mitochondria where it produces locales of BID [3]. In addition, the lipid composition of liposomes crucially modulated the ability of BID to permeabilize them, further substantiating a role for lipids in the action (and the targeting) of tBID [9]. In the second model, biochemical studies have substantiated a role for several mitochondrial proteins as receptors for tBID recruitment [10]. These include VDAC, as well as components of the mitochondrial protein import machinery, like TOM20, 22, 70 and 40 (reviewed in [11]), however, conclusive evidence for their role in this process is often lacking.

In a recent paper, Zaltsman *et al*. provide new important insights into the mechanism of recruitment of tBID on mitochondria. They define MTCH2/MIMP as a receptor for tBID on mitochondria and establish, using animal models, the importance of MTCH2/MIMP protein in Fas-induced hepatocellular apoptosis [1]. MTCH2/MIMP is a protein of the mitochondrial carrier family. In TNF-α treated cells, it resides in a 185 kDa large complex that comprises also tBID and BAX [12]. This evidence raised the hypothesis that MTCH2/MIMP could be involved in the mitochondrial apoptotic program but its role was not clear. Now, the paper of Zaltsman *et al.* closes this gap.

The first outstanding question that Zaltsman *et al.* tackled in their work was the submitochondrial localization of MTCH2. Being a member of the carrier superfamily, the natural prediction would be that it was located in the inner membrane. However, using three different biochemical approaches the authors clearly demonstrate that MTCH2/MIMP is in the outer membrane of the organelle. This result opened the possibility that MTCH2 participates in the steps of mitochondrial targeting of tBID during apoptosis. In order to verify this hypothesis, Zaltsman and coworkers assessed the role of MTCH2/MIMP *in vivo*. They first generated conventional knockouts, which were embryonically lethal when homozygous. They therefore created a conditional gene knockout mouse by using the Cre/loxP system. When MTCH2 is ablated, the cross-linkable complex tBID-MTCH2/MIMP is not detectable. Moreover, cells lacking MTCH2 are less sensitive to apoptosis induced by tBID and this defect is rescued by the reintroduction of MTCH2/MIMP. The reduced apoptosis is due to an impairment of tBID accumulation in mitochondria, following stimuli that converge on this BH3-only protein, including activation of Fas. Taken together, these results clearly demonstrated that MTCH2/MIMP protein is essential for the recruitment of tBID on mitochondria and it plays a fundamental role in the tBID-mediated cell death [1] (Figure 1).

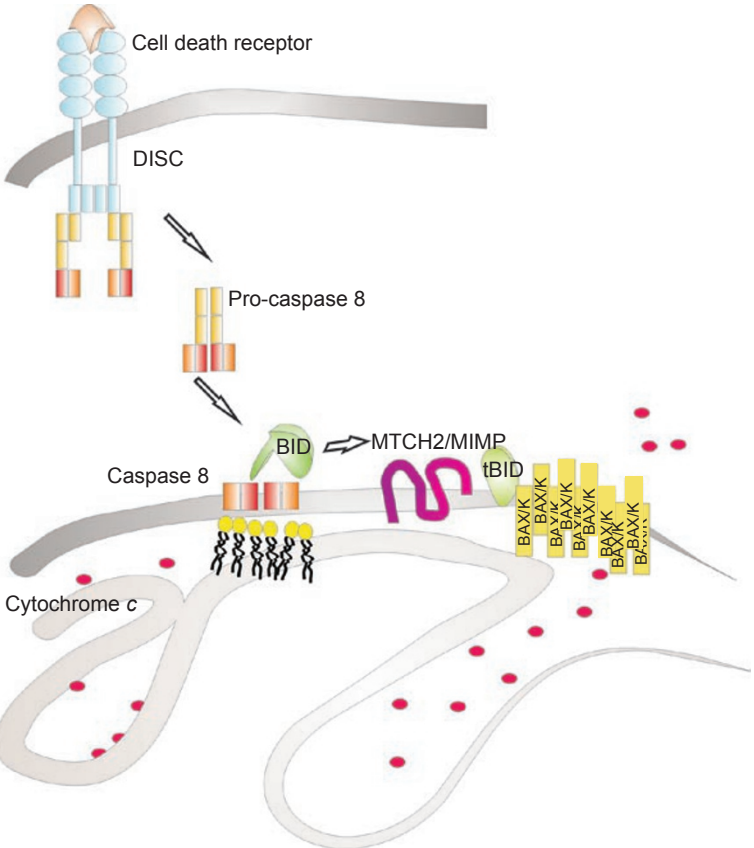

Figure 1 The recruitment of tBID on mitochondria is mediated by the novel target protein MTCH2/MIMP. The diagram depicts the sequence of events that occur in type II cells following an extrinsic death stimulus. Pro-caspase 8 binds to cardiolipin (yellow) on mitochondria where it undergoes self-proteolytic activation to cleave BID. The active tBID is then recruited on mitochondria by MTCH2/MIMP. This in turn leads to oligomerization of BAX/BAK and cytochrome *c* release.

It has been demonstrated *in vivo* that BID plays a key role in cell death induced by death-receptor ligands such as Fas ligand. In particular it exerts an important role in Fas ligand-induced apoptosis in hepatocytes [13]. Zaltsman and coworkers analyzed whether loss of MTCH2/MIMP that abolished *in vitro* recruitment of tBID on mitochondria could have significant effects on hepatocellular apoptosis *in vivo*.

They generated MTCH2/MIMP liver-specific knockout mice and assessed ther sensitivity to Fas. The liver-specific knockout animals show less liver injury and are more resistant to death than heterozygotes. In order to investigate the molecular mechanism of these effects, they analyzed the activation of caspases and the recruitment of tBID to mitochondria. The results clearly showed that in mice lacking MTCH2/MIMP in liver, caspase 8 was cleaved but the recruitment of tBID to mitochondria failed. This causes less activation of caspase 3 and consequently the hepatocites are less prone to apoptosis after Fas stimulation.

These results definitely confirmed that the MTCH2/MIMP protein plays a fundamental role in the recruitment of tBID to mitochondria and thus playing a key role in the Fas death pathway. Moreover, this paper contributes to clarify the basal mechanism of interaction of tBID with the mitochondria. Thanks to the paper of Zaltmans and co-workers, we now understand that the recruitment of tBID on mitochondria is at least partially orchestrated by a specific protein. However, it should be mentioned that the ablation of MTCH2 is not able *per se* to completely abrogate apoptosis, suggesting that other proteinaceous and/or lipidic receptors exist and play a role in the targeting of BID to mitochondria. Thus, it would be interesting to address if in cells lacking enzymes that participate in cardiolipin remodelling, like tafazzin [14], ablation of MTCH2 completely reduces it, substantiating a model in which lipids and proteins cooperate to target BID. Alternatively, one can envision a role for cardiolipin in targeting and/or assembly of MTCH2 in the outer membrane.

Another interesting link is that between MTCH2 and metabolism. MTCH2 recently emerged from a genetic screen as one of six new loci whose polymorphic variants are associated with increased body mass index. Among them, MTCH2 was the only one whose mRNA was not detected in the hypothalamus. This suggests that MTCH2 could play roles in the regulation of body mass in the periphery, as a regulator of energy expenditure. Apoptosis has been previously linked to metabolism when the BH3-only protein BAD was discovered to be a scaffold for enzymes of glucose metabolism on the surface of mitochondria, independently of its function in apoptosis [15]. However, in the case of MTCH2 it is not clear if the protein like BAD fulfils multiple functions in independent pathways, or if its role in apoptosis is key also for the regulation of body weight. In addition, it might be interesting to address if MTCH2 participates in the regulation of body weight by impacting on mitochondrial function. Studies capitalizing on the use of the conditional knockout animals generated by Zaltsman *et al.* will for sure help address these questions and place this protein in the broad context of integrated metabolism.

Acknowledgments

LS is a Senior Telethon Scientist of the Dulbecco-Telethon Institute and supported by The Italian Association for Cancer Research (AIRC), Telethon Italy.

References

1 Zaltsman Y, Shachnai L, Yivgi-Ohana N, *et al*. MTCH2/MIMP is a major facilitator of tBID recruitment to mitochondria. *Nat Cell Biol* 2010; **12**:553-562.
2 Danial NN, Korsmeyer SJ. Cell death: critical control points. *Cell* 2004; **116**:205-219.
3 Gonzalvez F, Schug ZT, Houtkooper RH, *et al*. Cardiolipin provides an essential activating platform for caspase-8 on mitochondria. *J Cell Biol* 2008; **183**:681-696.
4 Cipolat S, Rudka T, Hartmann D, *et al*. Mitochondrial rhomboid PARL regulates cytochrome *c* release during apoptosis via OPA1-dependent cristae remodeling. *Cell* 2006; **126**:163-175.
5 Frezza C, Cipolat S, Martins dB, *et al*. OPA1 controls apoptotic cristae remodeling independently from mitochondrial fusion. *Cell* 2006; **126**:177-189.
6 Scorrano L, Ashiya M, Buttle K, *et al*. A distinct pathway remodels mitochondrial cristae and mobilizes cytochrome *c* during apoptosis. *Dev Cell* 2002; **2**:55-67.
7 Zha J, Weiler S, Oh KJ, Wei MC, Korsmeyer SJ. Posttranslational N-myristoylation of BID as a molecular switch for targeting mitochondria and apoptosis. *Science* 2000; **290**:1761-1765.
8 Lutter M, Fang M, Luo X, *et al*. Cardiolipin provides specificity for targeting of tBid to mitochondria. *Nat Cell Biol* 2000; **2**:754-761.
9 Kuwana T, Mackey MR, Perkins G, *et al*. Bid, Bax, and lipids cooperate to form supramolecular openings in the outer mitochondrial membrane. *Cell* 2002; **111**:331-342.
10 Schafer B, Quispe J, Choudhary V, *et al*. Mitochondrial outer membrane proteins assist Bid in Bax-mediated lipidic pore formation. *Mol Biol Cell* 2009; **20**:2276-2285.
11 Ott M, Norberg E, Zhivotovsky B, Orrenius S. Mitochondrial targeting of tBid/Bax: a role for the TOM complex? *Cell Death Differ* 2009; **16**:1075-1082.
12 Grinberg M, Schwarz M, Zaltsman Y, *et al*. Mitochondrial carrier homolog 2 is a target of tBID in cells signaled to die by tumor necrosis factor alpha. *Mol Cell Biol* 2005; **25**:4579-4590.
13 Yin XM, Wang K, Gross A, *et al*. Bid-deficient mice are resistant to Fas-induced hepatocellular apoptosis. *Nature* 1999; **400**:886-891.
14 Xu Y, Malhotra A, Ren M, Schlame M. The enzymatic function of tafazzin. *J Biol Chem* 2006; **281**:39217-39224.
15 Danial NN, Gramm CF, Scorrano L, *et al*. BAD and glucokinase reside in a mitochondrial complex that integrates glycolysis and apoptosis. *Nature* 2003; **424**:952-956.