

Msp1: patrolling mitochondria for lost proteins

Ramanujan S Hegde

Intracellular protein localization is critical for establishing organelle identity, avoiding inappropriate interactions, and maintaining cellular function. An emerging mechanism for ensuring high-fidelity protein localization is the selective destruction of mislocalized copies. Two new papers find that the mitochondrial outer membrane is kept free of mislocalized proteins by Msp1 (Chen *et al*, 2014; Okreglak & Walter, 2014).

See also: YC Chen *et al* and V Okreglak & P Walter

A central problem in cell biology is to understand how the cell's approximately 10^9 protein molecules are distributed faithfully among its numerous membrane-bound compartments. The solution lies primarily in the multiple targeting and trafficking pathways that recognize signals in nascent proteins and direct them to their intended destinations. While these pathways are robust, minor populations of many proteins probably fail to be localized correctly (Levine *et al*, 2005). Mislocalization can be exacerbated in some situations (Kang *et al*, 2006) and even leads to pathologic consequences such as neurodegeneration (Chakrabarti & Hegde, 2009). Thus, attention has recently turned toward understanding the fate of mislocalized proteins.

The new studies focus on tail-anchored (TA) membrane proteins, defined topologically by a single transmembrane domain near the C-terminus. TA proteins of the cellular endomembrane system are first targeted and inserted into the endoplasmic reticulum (ER). The conserved "Guided Entry of TA protein" (GET) pathway carries out this task by recognizing nascent TA

proteins in the cytosol and delivering them to the ER membrane (Hegde & Keenan, 2011). Disruption of the GET pathway in yeast leads to TA proteins being partially mislocalized to the cytosol, and even inserted promiscuously into mitochondria (Fig 1). The consequences of this mislocalization are only now being studied.

In mammals, cytosolically mislocalized membrane proteins are recognized by the chaperone Bag6 (Hessa *et al*, 2011), and ubiquitinated by an associated ubiquitin ligase (Rodrigo-Brenni *et al*, 2014). Surprisingly, Bag6 had already been characterized as a metazoan-specific component of the TA insertion pathway (Mariappan *et al*, 2010). These discoveries suggested that localization pathways work in concert with degradation pathways to maximize fidelity of compartmentalization. Now, the Walter and Rutter laboratories build on this idea with their discovery of a surveillance pathway that clears mislocalized TA proteins from the mitochondrial outer membrane (Chen *et al*, 2014; Okreglak & Walter, 2014).

Both groups noticed that yeast GET pathway mutants show strong negative interactions with Msp1. Because Msp1 belongs to the AAA-ATPase family, members of which serve protein quality control functions (Wolf & Stolz, 2012), the authors suspected a role in clearing mistargeted TA proteins from mitochondria. This indeed proved to be the case: not only is Msp1 localized primarily to the mitochondrial outer membrane, but its deletion stabilized mitochondrially mislocalized TA proteins. An ATPase-deficient mutant of Msp1 was not only non-functional, but exerted a dominant-negative effect to stabilize mistargeted TA proteins on mitochondria. Excessive mistargeting, as occurs in GET deletion mutants, led to

altered mitochondrial morphology, a situation worsened by concomitant Msp1 deletion. Chen *et al* (2014) extended these observations to the mammalian system by showing that the Msp1 homolog ATAD1 plays a similar role. Thus, Msp1 is part of a conserved, ATPase-dependent pathway that eliminates mislocalized TA proteins from the mitochondrial outer membrane (Fig 1).

The biochemical function of Msp1 remains to be determined. The most attractive idea is that Msp1 directly mediates extraction from the membrane. Consistent with this, ATPase-deficient Msp1 was found associated with mislocalized, but not resident TA proteins (Chen *et al*, 2014). How Msp1-mediated dislocation might work, and whether this is coupled to downstream proteasome degradation pathways, is not known.

Another critical issue is how mislocalization is detected in the first place. In the case of Bag6, recognition occurs via exposed transmembrane domains which, if correctly localized, would be buried in a lipid bilayer. The Msp1 pathway appears to be more nuanced. An instructive finding is that Pex15, a peroxisomal TA protein, is not degraded from peroxisomes despite Msp1 being partially localized there. This suggests that the Msp1-TA protein interaction is contextual. For example, TA proteins in an inappropriate membrane may expose features that would normally be shielded by binding partners or a bilayer with different physical properties. The molecular basis of recognition and the factors involved in this decisive step are unresolved, but addressing this problem should be facilitated with the discovery of Msp1.

Finally, the physiologic importance of the Bag6 and Msp1 pathways for mislocalized

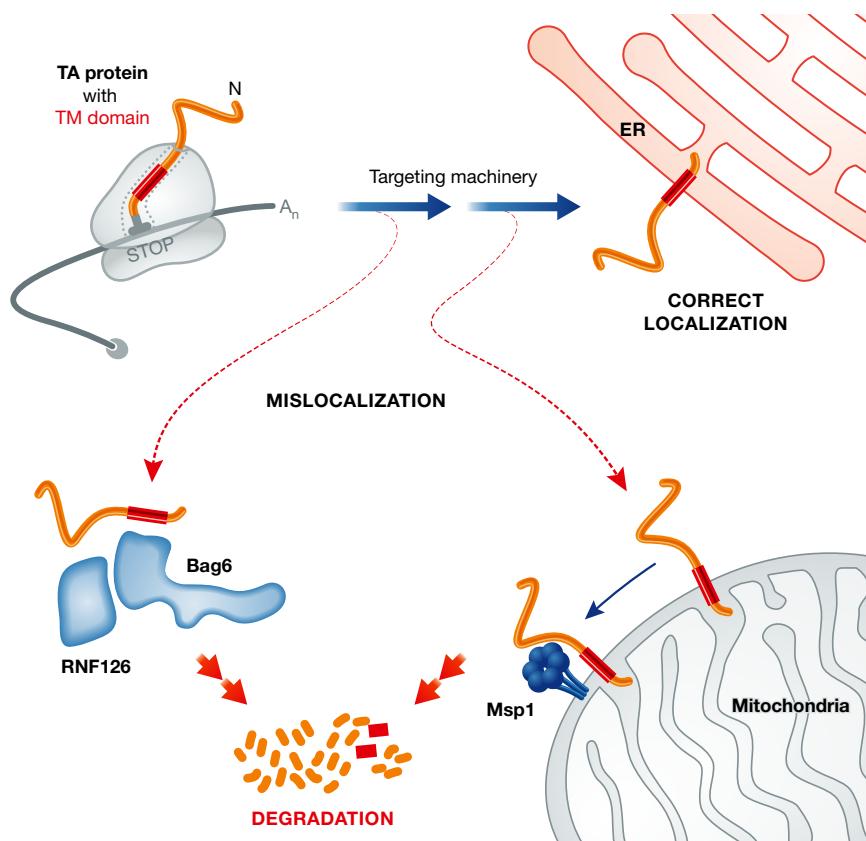


Figure 1. Pathways for mislocalized protein degradation.

Many tail-anchored (TA) proteins are destined for the endoplasmic reticulum. When everything works properly (top panel), a dedicated targeting and insertion machinery (not depicted) mediates correct TA protein localization to the ER membrane. Inefficiencies or failure of this targeting machinery results in mislocalization to the cytosol or mitochondria (lower panel), both of which contain factors that mediate degradation of mislocalized proteins. This includes the metazoan-specific chaperone Bag6 and its associated E3 ligase RNF126 in the cytosol, and the hexameric AAA-ATPase Msp1 in the outer mitochondrial membrane.

proteins merits investigation. Mislocalization affords opportunities for inappropriate interactions, aggregation, and other detrimental consequences (e.g. Chakrabarti & Hegde, 2009). Indeed, mitochondrial function is severely compromised upon excessive TA protein mislocalization (Chen *et al.*, 2014). Thus, previously unappreciated cleanup pathways operate tirelessly in the background to

maintain intracellular organization and avoid cellular pathology. Understanding the molecular basis of these pathways is an important future goal with implications for diseases of protein aggregation.

Conflict of interest

The author declares that he has no conflict of interest.

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