Review

Connection of Protein Transport and Organelle Contact Sites in Mitochondria

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Abstract

Mitochondrial biogenesis and function depend on the intensive exchange of molecules with other cellular compartments. The mitochondrial outer membrane plays a central role in this communication process. It is equipped with a number of specific protein machineries that enable the transport of proteins and metabolites. Furthermore, the outer membrane forms molecular contact sites with other cell organelles like the endoplasmic reticulum (ER), thus integrating mitochondrial function in cellular physiology. The best-studied mitochondrial organelle contact site, the ER–mitochondria encounter structure (ERMES) has been linked to many vital processes including mitochondrial division, inheritance, mitophagy, and phospholipid transport. Strikingly, ER–mitochondria contact sites are closely connected to outer membrane protein translocases. The translocase of the outer mitochondrial membrane (TOM) represents the general mitochondrial entry gate for precursor proteins that are synthesized on cytosolic ribosomes. The outer membrane also harbors the sorting and assembly machinery (SAM) that mediates membrane insertion of β-barrel proteins. Both of these essential protein translocases are functionally linked to ER–mitochondria contact sites. First, the SAM complex associates with an ERMES core component to promote assembly of the TOM complex. Second, several TOM components have been co-opted as ER–mitochondria tethers. We propose that protein import and organelle contact sites are linked to coordinate processes important for mitochondrial biogenesis.

Introduction

Mitochondria originate from an endosymbiotic event that occurred approximately 2 billion years ago when a eukaryotic ancestor cell incorporated a prokaryote related to α-proteobacteria [1,2]. In the course of evolution, the endosymbiont was transformed into a cell organelle, and almost its complete genetic information was transferred to the host nuclear genome. Nowadays, mitochondria still contain their own DNA, which, however, encodes only a small number of proteins: 8 proteins in baker's yeast *Saccharomyces cerevisiae* and 13 proteins in human mitochondria. Due to their endosymbiotic origin, mitochondria have two membranes, the outer and inner membrane, which give rise to two aqueous compartments: the mitochondrial matrix and the intermembrane space. The most prominent function of mitochondria is the generation of ATP for cellular metabolism via oxidative phosphorylation. The respiratory chain complexes generate a proton gradient across the inner membrane, which in turn drives the F_1F_O-ATP synthase. The respiratory chain complexes are enriched in invaginations of the inner membrane, termed cristae. In addition, several biosynthesis pathways are located within mitochondria including those for the biosynthesis of heme, amino acids, and lipids. Furthermore, iron–sulfur cluster formation takes place in the mitochondrial matrix. Defects in mitochondrial biogenesis and functions lead to neurodegenerative and myocardial diseases, revealing the central role of this organelle for health [3–5].

Mitochondrial function and biogenesis strictly depend on the uptake of proteins that are

synthesized as precursors on cytosolic ribosomes. The two surrounding membranes are equipped with a set of specific protein translocases that sort the precursor proteins into the mitochondrial subcompartments. In addition, the voltage-dependent anion channel (VDAC) of the outer membrane and specific transporters of the inner mitochondrial membrane allow the exchange of metabolites with the cytosol. In recent years, the discovery of a multitude of mitochondrial organelle contact sites has added a whole new layer of complexity to mitochondrial physiology. It emerged that mitochondria are firmly integrated into a cellular network for their biogenesis and function. They form molecular contacts to the endoplasmic reticulum (ER), vacuole, plasma membrane, and peroxisomes. These membrane contact sites are important for a number of functions including transport of lipids, maintenance of mitochondrial morphology, segregation of mitochondrial DNA, mitophagy, Ca^{2+} transfer, mitochondrial division, and inheritance [6–15].

In this review, we will focus on the functional connections between organelle contact sites and protein import into mitochondria. We propose that the crosstalk between these machineries serves to balance processes like maintenance of mitochondrial membrane architecture and lipid and protein biogenesis.

Mitochondrial Organelle Contact Sites

Due to their endosymbiotic origin, mitochondria are not embedded into the vesicular trafficking system. Instead, they form membrane contact sites to other cellular compartments including ER, plasma membrane, peroxisomes, and vacuolar membrane to ensure exchange of ions and lipids. Contact sites are defined as regions where two membranes come as close as 10–40 nm [14]. ER–mitochondria contact sites have been extensively studied. Already many years ago, electron microscopy revealed that mitochondria are closely located to the ER [16,17]. However, the proteins that tether both organelles remained undiscovered for decades. In the last years, several protein machineries that link mitochondria to the ER have been described in mammalian and yeast cells [12–15] (Table 1 and Fig. 1). The best-studied molecular tether between both organelles is the ER–mitochondria encounter structure (ERMES) in yeast. This evolutionarily ancient complex is not present in several eukaryotic lineages including animals [18,19]. The ERMES complex consists of four core subunits: The maintenance of mitochondrial morphology protein 1 (Mmm1) is anchored in the ER membrane. It interacts with the membrane-associated mitochondrial distribution and morphology protein 12 (Mdm12), which in turn is linked via Mdm34 to Mdm10. The β-barrel protein Mdm10 anchors the ERMES complex in the mitochondrial outer membrane (Fig. 1) [20–26]. The protein complex forms 2–10 foci per yeast cell [20,27,28]. The GTPase EF-hand protein of mitochondria (Gem1) associates with the ERMES complex and regulates the number of ERMES foci (Fig. 1), but it is not required for the structural integrity of the ERMES machinery [22–24].

In addition to its role as a molecular tether, the ERMES complex has been linked to several functions. ERMES subunits mark mitochondrial fission sites and are spatially localized to replicating mitochondrial DNA. The close vicinity between mitochondrial DNA and ER-associated mitochondrial division was reported to be important for the segregation of newly synthesized mitochondrial DNA to daughter mitochondria [29,30].

Table 1. Proteins and protein complexes involved in mitochondrial organelle contact sites and membrane architecture

Protein complexes and proteins	Proposed function	Localization
ER– mitochondria encounter structure (ERMES)	Tethers ER and mitochondria	ER/MOM
Mmm1	ER membrane anchor of the ERMES complex	
Mdm12	ER-associated protein that links Mmm1 to ERMES	
Mdm34	Mitochondria-associated protein that links Mdm10 to ERMES	
Mdm10	Mitochondrial membrane anchor of ERMES	
Gem1	Regulates number of ERMES foci	
Tom7	Controls segregation of Mdm10 between the ERMES and the SAM complex	
Vacuole and mitochondria patch (vCLAMP)	Molecular tether between vacuole and mitochondria	Vacuole/MOM
Vps39	Interacts with an unknown mitochondrial protein	
Ypt7	Rab GTPase, functions as vacuolar binding partner of Vps39	
ER membrane protein complex (EMC)	ER protein complex comprising eight subunits that interacts with mitochondrial Tom5	ER
Lam6	Regulates number of ERMES and vCLAMP foci and interacts with Tom70/Tom71	ER
Vps13	Localizes to vCLAMPs and nuclear–vacuolar junctions	Vacuole
Mcp1	Protein of unknown function, multicopy suppressor of Mdm10 mutants	MOM
Mitochondrial contact site and cristae organizing system (MICOS)	Mitochondrial inner membrane protein complex that forms contact sites to the mitochondrial outer membrane and promotes formation of crista junctions	MIM
Prohibitin	Oligomeric ring-like complex of prohibitin 1 and 2 that is involved in membrane organization	MIM