Biochimica et Biophysica Acta 1863 (2016) 2613–2623

Contents lists available at ScienceDirect

Biochimica et Biophysica Acta

journal homepage: www.elsevier.com/locate/bbamcr

Review

Orphan proteins of unknown function in the mitochondrial intermembrane space proteome: New pathways and metabolic cross-talk

Esther Nuebel, Phanee Manganas, Kostas Tokatlidis *

Institute of Molecular, Cell and Systems Biology, College of Medical, Veterinary and Life Sciences, University of Glasgow, UK

ARTICLE INFO

Article history:
Received 24 March 2016
Received in revised form 7 July 2016
Accepted 12 July 2016
Available online 15 July 2016

Keywords:
Mitochondria
Protein import
Intermembrane space
Redox signalling
Metabolism
Mitochondrial proteome

ABSTRACT

The mitochondrial intermembrane space (IMS) is involved in protein transport, lipid homeostasis and metal ion exchange, while further acting in signalling pathways such as apoptosis. Regulation of these processes involves protein modifications, as well as stress-induced import or release of proteins and other signalling molecules. Even though the IMS is the smallest sub-compartment of mitochondria, its redox state seems to be tightly regulated. However, the way in which this compartment participates in the cross-talk between the multiple organelles and the cytosol is far from understood. Here we focus on newly identified IMS proteins that may represent future challenges in mitochondrial research. We present an overview of the import pathways, the recently discovered new components of the IMS proteome and how these relate to key aspects of cell signalling and progress made in stem cell and cancer research.

© 2016 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/).

1. Introduction

Mitochondria are traditionally described in biochemistry textbooks as the power plants of a cell; although this is true, this limiting description underestimates the far reaching roles of this organelle in a variety of critical cellular processes unrelated to energy production. Mitochondria are key players in several cellular processes, including respiration, biosynthesis, apoptosis signalling and ion homeostasis. They are also generators of reactive oxygen species (ROS) which are themselves players in different signalling pathways [1,2]. These pathways share specific redox reactions [3], in which the mitochondrial oxidative defense system [4] contributes to maintain redox homeostasis.

Mitochondria have been implicated in several diseases, such as Parkinson's and Alzheimer's [5,6]. Additionally, their involvement in cancer [7,8], apoptosis signalling [9] and stem cell development [10] is under extensive research.

Modern techniques enabled the investigation of the internal structure and morphology of mitochondria and revealed a highly complex compartmentalisation [11,12]. The organelle is surrounded by a double membrane. This allows the assignment of the mitochondrial matrix (MM), the inner membrane (IM) the intermembrane space (IMS) and the outer membrane (OM). The inner membrane forms characteristic invaginations called cristae, which form an additional specific environment since they are separated from the inner boundary membrane by cristae junctions. Each different biosynthetic pathway can be assigned to a specific compartment, but the challenge is to dissect the communication and maintenance of the individual compartments. Part of this maintenance is to ensure proteostasis (folding, unfolding and degradation) to generate a homeostasis of the functional proteome and to clear mistargeted and damaged proteins. Every sub-mitochondrial compartment needs to control its redox milieu, which is interestingly highly different. The inner membrane separates the reducing matrix from the more oxidizing IMS [13]. For a long period of time, researchers focused on investigating the import pathways of the hundreds of nuclear encoded mitochondrial proteins. This culminated in the discovery of the major pathways that translocate proteins through the dedicated translocon complexes of the outer membrane (TOM), the inner membrane (TIM), as well as the MIA (Mitochondrial intermembrane space Import and Assembly) pathway, for the proteins that are targeted to the intermembrane space [14–17]. One of the big challenges of future research will be to investigate how the organelle communicates with the cytosol and the nucleus. The IMS with its specific redox environment and the 'controlled leakiness' of the outer membrane due to the presence of porins that allow the free diffusion of molecules less than 5 kDa, might harbour candidates which mediate the communication from signalling occurring inside the mitochondria towards other organelles, the cytosol and the nucleus. Of particular interest in this respect are newly identified IMS-resident proteins of known and unknown function and proteins with at least a dual

* Corresponding author.
E-mail address: Kostas.tokatlidis@glasgow.ac.uk (K. Tokatlidis).

http://dx.doi.org/10.1016/j.bbamcr.2016.07.004
0167-4889/© 2016 The Authors. Published by Elsevier B.V. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/).

localisation. For simplicity these proteins will be named 'orphans' not with the intension to classify necessarily their function as unknown but paying tribute to the fact that they have been assigned to the intermembrane space of mitochondria only recently. Such proteins could lead us to investigate new routes in mitochondrial research. In this review we aim to investigate new identified orphan proteins that are either soluble in the IMS or associated with the inner membrane and have a functional domain in the IMS. We interpret their features to address the possible pathways they might be involved in. This perspective will be complemented by an analysis of how signalling via metabolites and epigenetic modification may contribute to the intercellular cross-talk.

2. Mitochondrial import

Mitochondrial import has been studied extensively over the last decades [14,18–26]. There is still potential to unravel new pathways in addition to the well-established import routes. Protein import normally starts with binding of chaperones to the precursor located in the cytosol, followed by binding to import receptors located at the outer mitochondrial membrane. The precursors then pass through the TOM complex, which, in yeast, is made up by the proteins Tom20, 22, 70, the pore forming Tom40 protein and small components named Tom 5, 6, and 7. The precursor is subsequently guided via the TIM receptors Tim50 and Tim23 and engages the TIM pore which is formed by the Tim17 and 23 proteins. The import process of matrix proteins is driven to completion by an ATP consuming step, which involves Tim44, mitochondrial Hsp70 and the nucleotide exchange factor GrpE (Mge1). If a targeting signal is present, this is cleaved by an internal protease, which subsequently allows the refolding of the protein [21,27]. The sorting and assembly (SAM) pathway for outer membrane proteins as well as the second TIM pathway and the MIA dependent import into the IMS are distinct translocons that allow a compartment-specific localisation of nuclear encoded proteins.

3. Import into the IMS

Here, we focus on two main pathways for import into the IMS which have been well characterised. The first class of proteins follows the same import route as matrix proteins but a hydrophobic so called "stop-transfer" sequence causes an arrest of the precursor within the TIM23 complex, cleavage of the N-terminal presequence peptide by the matrix peptidase MPP, followed by a second proteolytic cleavage of the stop-transfer sequence in the IMS and release of the mature protein into this compartment [28]. Different peptidases are known to participate in this second cleavage event depending on the substrate protein, e.g. cytochrome b_2 and Mgm1 are cleaved by Imp1 and Pcp1, respectively. A second class of IMS proteins are the ones that acquire disulfide bonds during their biogenesis. A dedicated machinery oxidizes these proteins as part of their import process into the IMS [29–33]. As mentioned earlier, the redox status of the IMS is oxidizing and is required for this pathway. It is likely that the IMS of mitochondria has maintained the oxidizing environment of its corresponding bacterial compartment, the periplasm, during the endosymbiotic process. The redox potential of the intermembrane space is around −225 mV, which is more oxidizing than that of the cytosol at −290 mV [13]. However, Kojer and co-workers [34]suggested (using a new generation of roGFP-based redox-sensors) that the levels of GSH are maintained in the IMS through a porin-mediated diffusion from the cytosol. The identification of the enzymes, Mia40 [30] and Erv1, which perform regulated transfer of disulfide bonds to substrate proteins, was a substantial advance for the intermembrane space oxidative pathway [35].

The mechanism for protein import into the mitochondrial intermembrane space starts with the reduced and, therefore, disulfide bond free substrates entering via the OM. Disulfide bonds are introduced by Mia40, and electrons are transferred from Mia40, to oxidized Erv1, and finally to oxygen produced by the respiratory chain via cytochrome c and complex IV [36]. The final electron acceptor under anaerobic conditions remains elusive. The introduction of a disulfide bond increases the tertiary structure of the substrate and therefore traps the protein in the IMS.

The targeting sequence for this class of proteins is called Intermembrane space Targeting Signal (ITS) or mitochondrial IMS-sorting signal (MISS) and is found additionally to the CX_nC cysteine motifs (e.g. the CX_9C motif in yCox17) [37,38]. The ITS is characterised by some key properties: (i) it can function upstream or downstream of the cysteine which interacts with Mia40, also called the docking cysteine; (ii) it is sufficient for crossing the outer membrane and even for targeting non mitochondrial proteins; (iii) it forms an amphipathic helix with hydrophobic residues facing the side of the docking cysteine and dispensable charged residues on the other side and (iv) its fit is complementary to the substrate cleft of Mia40 via hydrophobic interactions [39].

4. Proteome of the IMS

The complexity of the proteome supersedes that of the genome, due to alternative splicing events and post-translational modifications, such as phosphorylation or methylation. Here, we want to focus on the recently undertaken approaches to investigate the proteome of the smallest sub compartment of mitochondria, the intermembrane space. The intermembrane space proteome requires careful analysis since the last decades of research have pointed out the importance of this sub compartment in processes such as (i) protein transport, (ii) lipid transport, (iii) regulation and assembly of the respiratory chain, (iv) regulation of redox processes, (v) coordination of apoptosis and (vi) metal homeostasis. It is under investigation to unravel the so far unknown pathways the IMS proteome might be involved in [40]. The investigation of the IMS holds potential to unravel players which are involved in cysteine oxidation due to its oxidizing environment, unlike the prevention of cysteinereduction in other cell compartments like the cytosol [41]. It is also tempting to speculate about the presence of a very tightly regulated redox sensing mechanism which will help to adjust the metabolism to a given stressor. In cases of stress, induced from a variety of sources including chemicals, metals, or particular diseases, the cell needs to make the decision whether to undergo programmed cell death or repair. In the case of apoptosis, it is widely recognised that the IMS releases part of its proteome [42]. The molecules which might eventually be targeted to the IMS only under specific stress conditions have not yet been investigated in detail. Previous studies could identify and verify the localisation of 31 IMS proteins in yeast [43] and 23 IMS proteins in human mitochondria [44]. More recent investigations have been able to detect a number of new IMS orphans in both organisms. More specifically, these efforts were performed using the established yeast model system *S. cerevisiae* [43] and, more recently, human cells. Hung and co-workers were able to extend the current human IMS proteome to a protein count of 127 [45].

5. New identified orphans of the IMS

The main challenge in proteomics is to achieve high purity of subcellular regions without contaminations from distinct organelles. This is not a trivial task, as contact sites between organelles are frequently part of their normal function and separating them in homogeneous and pure fractions is very difficult. Since this technical challenge limits our understanding of pathways and mechanisms in living cells, researchers have devoted substantial efforts to alleviate this problem. In the recent literature, two attempts have been reported to create a full inventory of the IMS proteome. Vögtle et al. induced BAX mediated release of the IMS proteome to map all IMS located candidates in *S. cerevisiae* cells [43]. More recently, another group developed a very intriguing method, where they used an engineered ascorbate peroxidase (APEX), which was genetically targeted to the IMS. Upon addition

Download English Version:

https://daneshyari.com/en/article/5508682

Download Persian Version:

https://daneshyari.com/article/5508682

Daneshyari.com