Friday, March 22, Session VI

L21

NEW SCRAMBLASES WITH MOONLIGHTING FUNCTIONS IDENTIFIED IN MITOCHONDRIA

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Mitochondria are the essential powerhouses of eukaryotic cells, and their optimal function depends on a specific phospholipid composition in their two membranes. While some of these lipids are supplied by the endoplasmic reticulum, the mechanism that facilitates their transfer across the outer mitochondrial membrane has been elusive. Our research has identified novel scrambling proteins responsible for lipid transport across the outer mitochondrial membrane.

The first identified protein is the voltage-dependent anion channel (VDAC), an abundant component of the mitochondrial outer membrane [1]. Although VDAC is widely recognized for its ion channel activity, our research has revealed its previously unknown role as a phospholipid scramblase. This novel function was demonstrated with in vitro experiments using yeast mitochondria and reconstituted vesicles, and is further supported by coarse-grained molecular dynamics simulations of VDAC [2]. These simulations have elucidated the mechanism of lipid scrambling, which occurs at specific interface between VDAC dimers (Figure 1a). This interface contains polar residues that create significant water defects and thin the lipid bilayer, facilitating lipid scrambling. VDAC, with its beta-barrel transmembrane structure, introduces a new class of phospholipid scramblases [2], which are distinct from previously identified scramblases with alpha-helical transmembrane structures.

The second identified protein, MTCH2, is also located in the mitochondrial outer membrane and has previously been recognized as an insertase [3]. Unlike VDAC, MTCH2 is characterized as a helical transmembrane protein similar to previously identified scramblases. MTCH2 has a hydrophilic groove in the transmembrane region that locally thins the hydrophobic core of the membrane and facilitates the lipid scrambling (Figure 1b), similarly to VDAC [4]. Molecular dynamics simulations, both coarse-grained and atomistic, were used to demonstrate that the groove is responsible for lowering the free energy barrier for lipid movement across the membrane. The scrambling rate observed for MTCH2 is comparable to that of VDAC (see Figure 1c), suggesting its potential complementary role in mitochondrial lipid transport. Although VDAC and MTCH2 have distinct structural features, they share a common mechanism of facilitating lipid transport by locally thinning the membrane [4].

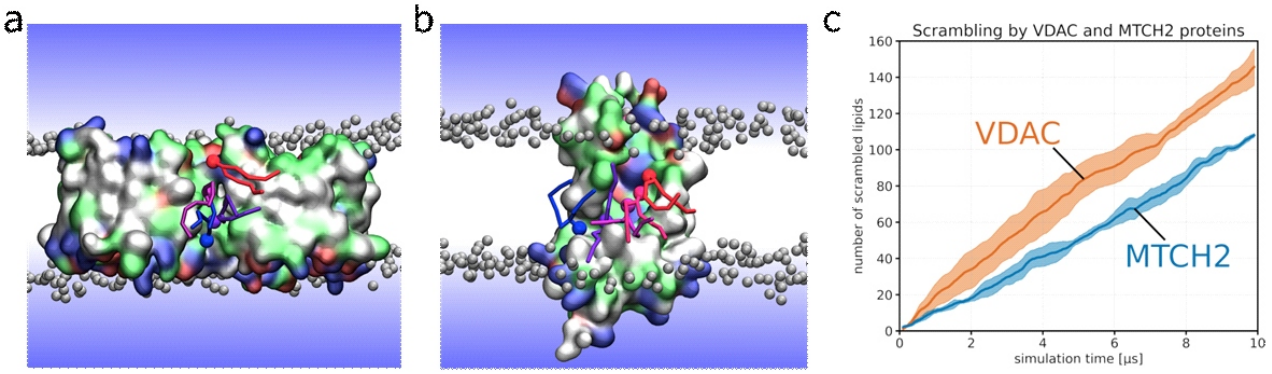

Figure 1. Lipid scrambling by VDAC and MTCH2 proteins. Snapshots of VDAC dimer a) and MTCH2 b) from coarse grained simulations depict phospholipids translocating between membrane leaflets along the scrambling pathway at the protein. The translocating lipids are colored in red, purple, and blue. Proteins are shown as a molecular surface, with colors indicating the character of its residues (hydrophilic = green, hydrophobic = white, positively charged = blue, negatively charged = red). Lipids of bulk membrane are depicted as gray beads (phosphate groups) with hydrophobic tails omitted for clarity. Water is represented only schematically as a blue gradient. c) Scrambling rate of VDAC dimer and MTCH2 proteins in a POPC membrane are shown as the number of "scrambled lipids" in time. Lipid was considered scrambled when it was present in the opposite leaflet than its original one.

These discoveries represent a significant advancement in our understanding of lipid transport in mitochondria. In addition, the dual functionality of these proteins highlights the complexity of mitochondrial biology and provides opportunities for further research in membrane dynamics and cellular metabolism.

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THREE EPOCHS OF NASCENT PROTEIN TRANSLOCATION THROUGH THE RIBOSOME EXIT TUNNEL

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All proteins in living organisms are produced in ribosomes that facilitate the translation of genetic information into a sequence of amino-acid residues. During translation, the ribosome undergoes stages of initialization, elongation, termination, and recycling. In fact, peptide bonds are formed only during the elongation stage, which comprises periodic association of transfer RNAs and multiple auxiliary proteins to the ribosome, and adding an amino acid to the nascent polypeptide one at a time.

Protein spends a considerable amount of time attached to the ribosome. In this context, we conceptually divide the con-translational lifespan of protein into three epochs. We define the epochs on the basis of the position of the N-terminus of the nascent polypeptide within the ribosome exit tunnel and the context of the catalytic center.

In the first epoch, the N-terminus of the nascent protein travels from the catalytic site of the ribosome towards the tunnel exit. During the second epoch, the N-terminal part of the nascent protein remains outside the ribosome and typically undergoes co-translational folding. In the third epoch, the C-terminus is cleaved off the tRNA, escaping from the ribosome exit tunnel.

In the talk, we argue that nascent proteins experience a variety of forces that determine how they translocate through the tunnel and interact with the tunnel walls. We review current knowledge about translocation and identify several gaps in our understanding of the birth of proteins.

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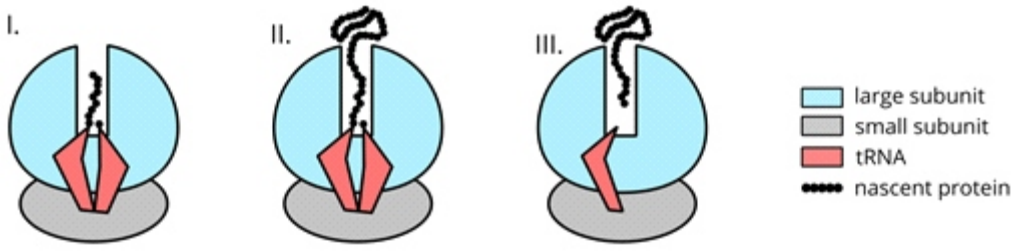

Figure 1: A schematic representation of the three epochs of nascent protein translocation