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Impact of N-terminal Acetylation on the structure and molecular interactions of the "GRASP65 homolog protein 1"from Saccharomyces cerevisiae

SILVA, Marília Darini de Oliveira da[1]; MENDES, Luis Felipe Santos[1]; SILVA, Mayra Tamires Santos[1]

mayra.tamires@ifsc.usp.br

[1]Instituto de Física de São Carlos – USP

The Golgi apparatus, a vital organelle in eukaryotic cells, plays crucial roles in modifying, storing, and transporting proteins and lipids. Its distinctive architecture is predominantly shaped by Golgi Reassembly and Stacking Proteins (GRASP) and Golgins. (1) The importance of these proteins in the Golgi organization is widely recognized, and recent advances in understanding their biochemical and biophysical properties have shed new light on the Golgi organization, presenting a perspective that challenges traditional notions. (2) However, the precise understanding of how they modulate the compartments of this apparatus to polarize the direction of protein secretion, especially at the molecular level, remains obscure. The membrane anchoring of GRASPs involves a crucial post-translational modification at the N-terminal. However, studies on these proteins often use heterologous expressions in prokaryotic systems, which cannot perform such necessary modifications. In this work, we proposed to elucidate this enigma by establishing a protocol to investigate the role of N-terminal acetylation in the molecular interactions of the homologous GRASP protein from Saccharomyces cerevisiae (ScGRASP), also called GRASP65 homolog protein 1, in biological membrane models. The N-terminal acetylation of this protein was enabled by the catalytic properties of the NatC complex, formed by the Naa30, Naa35, and Naa38 subunits. (3) Through circular dichroism and calorimetric studies, combined with the use of membrane mimetics, we detected a disorder-to-order transition in the N-terminus that was strongly dependent on the N-terminal acetylation and the presence of negative phospholipids on the membrane surface. This structural transition is responsible for forming an amphipathic helix, which is shown to be essential for the membrane anchoring properties of ScGRASP. We aim, with this, to provide experimental data that can significantly enhance our understanding of the molecular mechanisms governing the architecture of the Golgi apparatus.

Palavras-chave: Biological membranes; Spectroscopy; Biochemistry.

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