Investigating Site-Specific Acetylation of S-OPA1: Implications for GTPase Activity and Oligomeric Assembly

Mitochondrial dynamics are regulated by fission and fusion reactions, governed by proteins and lipids organized spatially within the mitochondria[1,2]. The dynamin-related GTPase optic Atrophy 1 (OPA1), plays a critical role in mitochondrial inner membrane fusion and maintaining the architecture of cristae[3,4]. Recent studies suggest that post-translational modifications, such as lysine acetylation, significantly influence the activity of mitochondrial proteins[5], yet the specific effects on S-OPA1 remain poorly understood. In this study, we used acetylation mimetic mutations to investigate the impact of site-specific lysine acetylation on s-OPA1 function. Using a combination of in silico and functional assays, we identified key lysine residues on s-OPA1 that upon acetylation/deacetylation influence protein stability, GTPase activity, and higher-order oligomeric assembly. Our findings reveal that acetylation at K328 and deacetylation at K342 in G domain increases the GTPase activity of the protein upon binding to lipid membranes whereas deacetylation at K772 completely abolishes membrane binding-induced GTPase activity. Using negative staining transmission electron microscopy, we showed that acetylation of lysine on s-OPA1 does not impact the ability of the protein to bind and tubulate liposomes, but affects higher-order oligomeric assembly. These insights enhance our understanding of site-specific acetylation in regulating s-OPA1 activity and highlight its potential implication in mitochondrial-related diseases.

Keywords: Mitochondrial Dynamics, OPA1, Acetylation, Mimetic mutations, oligomeric assembly

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