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DynA may have a role in membrane fusion events during cytokinesis



Members of the dynamin superfamily of GTPases are known to be mediators of membrane remodelling processes in eukaryotic cells. Many bacterial genomes also contain genes encoding dynamin-like proteins, but their function has remained unclear. Writing in *Molecular Microbiology*, Bürmann *et al.* now show that DynA, a dynamin-like protein found in *Bacillus subtilis*, localizes to the septum of dividing cells and can mediate nucleotide-independent membrane fusion *in vitro*.

Some bacterial genomes contain multiple genes encoding dynamin-like proteins, with two genes often occurring in tandem. In *B. subtilis*, these tandem genes have become fused, resulting in a protein (DynA) that contains two dynamin-like GTPase domains. To investigate the possible function of this protein, the authors expressed wild-type and mutant forms of DynA in *Escherichia coli* and then purified them for biochemical analyses. Using a cross-linking approach, the authors confirmed that DynA binds GTP. Mutation of the P-loop (a central

motif in the GTPase domain) in either of the DynA GTPase domains (K65A or K625A) reduced GTP binding, but only a double mutant (K65A and K625A) abolished GTP binding completely. However, GTPase activity was lost in both single P-loop mutants as well as in the double mutant, suggesting that GTP binding by both GTPase domains of DynA is needed to complete the hydrolysis cycle.

Despite this intrinsic GTPase activity, DynA exhibited nucleotide-independent membrane binding *in vitro*, in contrast to many other members of the dynamin superfamily, and was also able to tether liposomes into large clusters. Furthermore, DynA led to fusion of the liposomes, as shown in FRET-based assays in which liposomes containing 4-nitrobenzo-2-oxa-1,3-diazole (NBD) and rhodamine fluorophors were mixed with unlabelled liposomes in the presence of wild-type DynA or each of the P-loop mutants. P-loop mutants were able to induce liposome fusion, indicating that this process is nucleotide independent,

but fusion did require the presence of magnesium ions. Finally, the authors observed that a GFP-tagged version of DynA preferentially localized to the sites of septation in dividing *B. subtilis* cells. Interestingly, in cells lacking MinJ (an important structural component of the divisome), DynA became dispersed along the entire cell membrane rather than localizing to the septum.

Together, these findings suggest that DynA may have a role in membrane fusion events during cytokinesis. However, the authors note that a Δ *dynA* *B. subtilis* strain does not exhibit any morphological difference when compared to wild type, suggesting either that there is a redundant system that can perform the same function as DynA or that the activity of DynA is only required under specific environmental conditions.

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