BACTERIAL PHYSIOLOGY

Energizing the spore coat

Spores formed by *Bacillus subtilis* are encased in a complex multilayered coat that comprises more than 50 different proteins. Ramamurthi and Losick have published a study in *Molecular Cell* which shows that ATP hydrolysis by the morphogenic coat protein SpoIVA, which enables the self-assembly of SpoIVA polymers, is of central importance in spore-coat formation.

Spore formation occurs in a sporangium, which consists of a mother cell that contains an inner cell (forespore) that eventually forms the mature spore. Spore-coat components are synthesized inside the mother cell and deposited on the outside of the forespore. The basement layer of this coat is formed by two proteins; SpoVM interacts directly with the forespore membrane and tethers SpoIVA, which recruits the various factors that are involved in formation of the spore coat. Deletion of SpoIVA results in the formation of coats that are not tethered to the forespore, but are instead free in the cytosol of the mother cell.

Bioinformatic analysis of SpoIVA revealed a highly conserved seven-amino-acid nucleotide-binding Walker A motif. Mutation of a single lysine in this motif was sufficient to prevent SpoIVA from localizing to the forespore membrane, causing a complete block in sporulation. In addition, although SpoVM was still able to localize correctly, recruitment of other coat proteins, such as CotE, was altered, which shows that an intact SpoIVA Walker A motif is important for spore-coat formation. The authors found that SpoIVA bound ATP with strong affinity, but hydrolysed the ATP slowly, confirming that the SpoIVA Walker A motif functions in a classical nucleotide-dependent fashion.

Why does SpoIVA need to hydrolyse ATP? To answer this question the interaction between SpoIVA molecules was probed. Using an affinity purification approach of proteins from cells that expressed two forms of tagged SpoIVA, the authors found that mutation of the Walker A motif was sufficient to block interactions between SpoIVA molecules. Further analysis revealed that purified SpoIVA could only form high-molecular-weight oligomeric complexes in the presence of ATP. Surprisingly, the oligomeric complexes aggregated in the reaction tube, forming strings that were composed of bundles of SpoIVA filaments. This observation suggests that in addition to the linear interactions required to form polymers, SpoIVA can interact *in trans* with adjacent SpoIVA molecules, thereby enabling the filaments to form bundles, a property that would be useful during formation of the spore-coat basement layer.

This self-assembly mechanism, which is driven by nucleotide hydrolysis, is likely to be important for formation of the spore coat. Further work is needed to characterize the intermolecular interactions enabled by ATP hydrolysis, which will provide a framework for better understanding how the spore coat is deposited onto the outer forespore membrane.

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