Actin-binding Proteins

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The goal of the Robinson laboratory is to investigate the structural and functional aspects of a range of actin-binding proteins. From these data we aim to gain insight into processes that harness actin polymerization for cellular functions.

We have determined the crystal structures of proteins, or protein motifs, that fall into the classification of being “structurally disordered”. While these proteins may be “structurally disordered” in isolation they adopt distinct conformations on interacting with their binding partners. One such example is that of the WH2/thymosin beta family, or families of proteins. These molecules, in general, bind to ATP-actin monomers. However, in some situations they relinquish the bound ATP-actin to actin filament nucleation machineries, such as the arp2/3 complex, or indeed interact with actin protomers with a filament.

The WH2/thymosin beta family proteins comprise a linear stretch of around twenty amino acids. The N-terminal portion forms and amphipathic helix that binds to between actin subdomains one and three, while the C-terminal portion is extended and lies across the external surface of actin. This C-terminal region includes an LKK(T/V) motif that is the distinguishing feature of this family of proteins. The thymosin beta proteins differ from WH2 proteins through having a further twenty amino acids that form a mirror image of the first twenty amino acids, in forming an extended region followed by an amphipathic helix. This helix binds between actin subdomains two and four at the opposite end of the actin protomer to that of the N-terminal helix (Fig. 1).

Fig. 1: Thymosin beta interactions with actin

These differences between the thymosin beta structures and WH2 structures are providing answers as to how some motifs are able to sequester actin monomers whilst others can participate in the actin filament elongation process.

A second family of “structural disordered” motifs are those that bind to capping protein. Capping protein is a heterodimer of symmetrically related halves, which forms the universal barbed end cap for actin filaments. The barbed end of an actin filament is the polymerization-active end in cells. Free barbed ends are highly regulated to allow explosive polymerization to be utilized for processes that require polymerization force, such as membrane protrusion. This requires, that more generally, the barbed ends should be capped so as not to deplete the acin monomer pool in non-productive polymerization.

Removal of capping protein from an actin filament is one method, at least *in vitro*, of restoring a capped filament to a polymerization competence. A family of such proteins are able to wrap around capping protein and alter its actin binding dynamics (Fig. 2).

Fig. 2: Capping protein interacting motif

Taken together these structures are providing insights into actin dynamics and elucidating the roles of “structurally disordered” regions of proteins.