

Role of Myosin 1b in actin network assembly by in vitro approaches

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The actin cytoskeleton is the dynamic architecture of living cells made of several structurally and functionally distinct arrays of actin filaments. Establishment and maintenance of a polarized network of growing filaments require links between the barbed ends of filaments and the membrane. Actin-binding and membrane-associated proteins link the growing barbed ends of filaments to the membrane to control both their polarity and dynamics. Cell shape changes require forces that are generated on membranes by the combined action of actin filament polymerization and myosin motor activity. Despite a wealth of information on myosin activity, the role played by these proteins of myosin 1 family on the architecture and dynamics of cellular actin networks remain unclear.

Myosin 1b (Myo1b) is a single-headed membrane-associated motor that bind actin filaments. It comprises an N-terminal motor domain coupled to a C-terminal tail homology 1 domain (containing a lipid-binding domain) by a neck region that binds Calmodulin. A new mechanism can be envisioned for Myo1b-induced membrane deformation. Myo1b could act as an active tether between polymerizing actin filaments and the membrane. The force generated by actin polymerization could be transduced by Myo1b into membrane tubulation. If the actin filaments are nucleated by formins and if myosins help orientating elongated filaments, a polarized actin network could be observed. The polarization will depend on the equilibrium between processive elongation by formins and sliding of the filaments by myosins motor activity. Alternatively Myo 1b may control the arp2/3 actin network required for the membrane deformation.

The goal of this project is to determine the mechanism by which membrane-associated Myo1b activity controls the elongation and orientation of growing actin filaments to shape a polarized network of actin filaments that can deform the membrane. To this aim, we developed, new *in vitro* microscopy assays with pure proteins.