Impact of mechanics on actin disassembly by ADF/cofilin

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In the regulation of the actin cytoskeleton, disassembling networks is as important as assembling them. At the centre of the disassembling machinery is the family of actin-binding proteins ADF/cofilin. ADF/cofilin binds to the side of filaments forming domains that alter the conformation of actin monomers, increasing the filament helical pitch.

This change in structure leads to the severing of filaments thus accelerating the disassembly of networks. However how these shorter filaments then fully depolymerise to G-actin is not understood. A second important question is how ADF/cofilin targets specific actin filaments to disassemble. While biochemistry certainly plays an important role, it has also been proposed that mechanical stress could tune the effects of ADF/cofilin.

To tackle these questions, we use a microfluidic setup in which actin filaments are attached to the surface by one end or several points along their length. Through protein labelling and TIRF microscopy, we measure the depolymerisation dynamics at each end, ADF/cofilin binding and induced severing.

We discovered that ADF/cofilin not only accelerates the depolymerisation at the pointed end but also allows filaments to disassemble from their barbed end: in cells while ATP-G-actin should bind the barbed end to elongate filaments, ADF/cofilin synergies with Capping Protein to saturate filaments and put their barbed end into an unstoppable depolymerising state.

We then assessed the effect of tension, bending and twisting, mechanical stresses that occur in actin networks. While we observed no effect of tension, we found that filaments that cannot twist freely break faster: upon binding, ADF/cofilin puts filaments in an under-twisted state that promotes their severing.

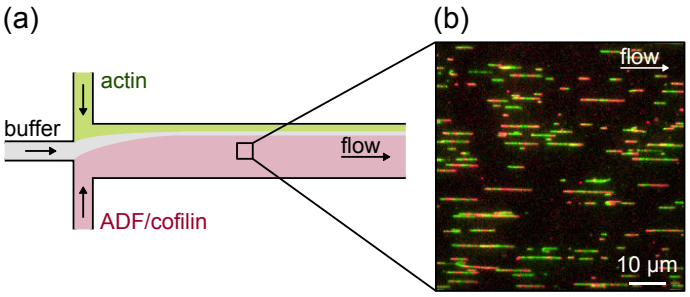

Figure 1: Microfluidic setup. (a) Sketch of the channel, solutions are injected from the three inlets at controlled flow rates. (b) Typical field of view. Actin filaments (green), covered with ADF/cofilin domains (red), are aligned with the flow.