

# **Generation of stress fibers depends on mechanosensitive actin filament assembly and disassembly**

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**Ventral stress fibers are the major contractile actomyosin bundles in most non-muscle cells, where they have important role in adhesion, morphogenesis and migration. Stress fibers are mechanosensitive structures, but how tensile forces control actin dynamics to drive their assembly and alignment have remained elusive. We show that arcs, which serve as precursors of contractile ventral stress fibers, fuse with each other during centripetal flow to form thick actomyosin bundles that apply tension to focal adhesions located at their ends. Importantly, this tensile force inhibits vectorial actin polymerization at focal adhesions through VASP phosphorylation to halt stress fiber elongation, and thus ensures their proper contractility. Stress fiber maturation additionally requires ADF/cofilin-mediated disassembly of non-contractile stress fibers, whereas contractile fibers are protected from filament severing. These data reveal that myosin-derived tension precisely controls both actin filament assembly and disassembly to ensure generation and proper alignment of contractile stress fibers in migrating cells.**