Poster Presentation

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Molecular Analysis of a G-actin Sensor

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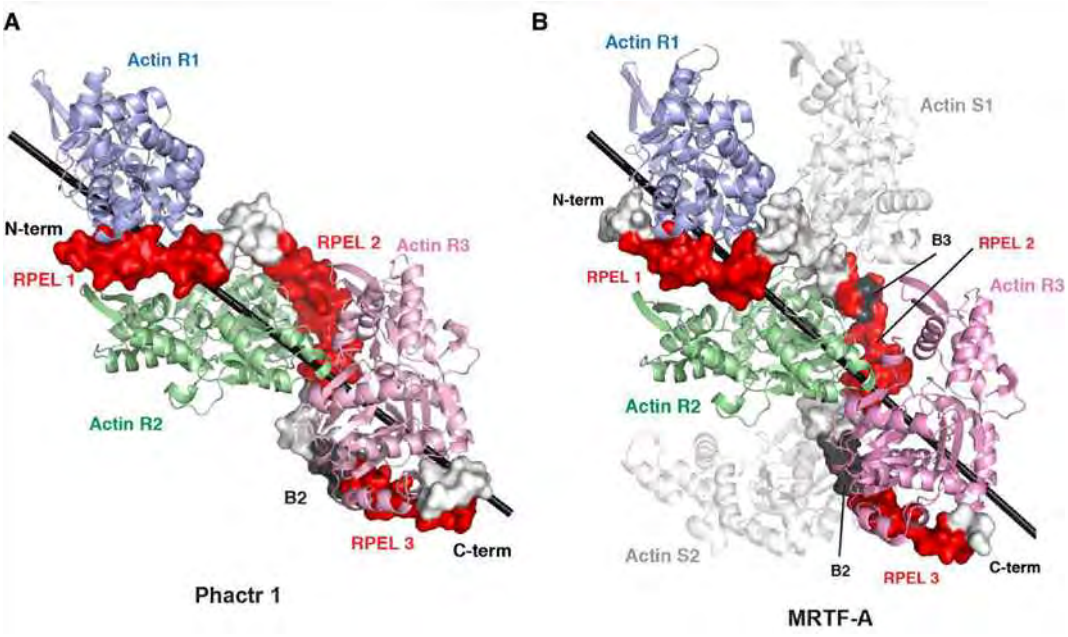
Actin dynamics control many aspects of cell shape and cell motility through regulatory interactions with a large variety of actin-binding proteins. Signalling to these actin regulators frequently involves a Rho GTPase-stimulated pathway that leads to a dramatic fluctuation in the levels of monomeric actin (G-actin) following polymerisation to F-actin. Recent studies have identified a molecular G-actin sensor called the RPEL domain that links RPEL-containing proteins and their subcellular localisation to actin dynamics. The RPEL domain contains a tandem array of typically three RPEL motifs, each of which is competent to bind a G-actin molecule [1]. The domain is present in two otherwise unrelated protein families; the MRTF family of serum response factor (SRF) transcriptional co-activator proteins and the Phactr family of actin and PP1 phosphatase-binding proteins. We have begun to investigate how the RPEL domain operates in both of these protein contexts and how it modulates subcellular localisation, transcriptional regulation and actomyosin contractility. To define the molecular basis for the sensor we have reconstituted pentameric and trimeric G-actin complexes with the RPEL domain from both MRTF-A and Phactr and used crystallography to reveal discrete supramolecular assemblies with repetitive arrangements of the G-actin subunits around the "crankshaft"-shaped RPEL domain [2,3]. These arrangements are quite different from F-actin intermolecular contacts and are quite unexpected. Our crystal structures reveal cooperative loading of G-actin onto the RPEL domain that we show by several cell-based reporter assays to be of functional importance. These structures explain how G-actin interaction alters the subcellular localisation of both MRTF-A and Phactr by inhibiting nuclear import through competing with importin alpha-beta binding [2,3].

[1] *S. Mouilleron, S. Guettler, CA. Langer, et al, 2008, EMBO J., 27, 1-11,* ***[2]*** *S. Mouilleron, CA. Langer, S. Guettler, et al, 2011, Sci. Signal., 4, ra40,* ***[3]*** *S. Mouilleron, M. Wiezlak, N. O'Reilly, et al, 2012, Structure, 20, 1960-1970*



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