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Double JMY: making actin fast

David W. Roadcap and James E. Bear^{*}

Lineberger Comprehensive Cancer Center and Dept. of Cell & Developmental Biology, University of North Carolina-Chapel Hill, Chapel Hill, NC, USA, 27599

Abstract

Assembly of actin networks is dependent on nucleation promoting factors. A new study identifies JMY as a protein containing two separate nucleation promoting activities. JMY shuttles between the nucleus and the cytoplasm and promotes cell migration. These observations indicate that JMY is an important factor controlling actin dynamics in motile cells.

Actin filaments provide the structural basis for much of cell motility, and therefore are critical to numerous physiological processes such as morphogenesis, wound healing, and immune response. Abnormal cell migration also plays a role in disease states such as autoimmune disorders and metastatic cancer. To better understand these processes, comprehensive knowledge about the mechanisms for promoting, inhibiting, and regulating actin dynamics is required. The first, rate-limiting step in forming actin filaments is the *de novo* nucleation of actin filaments from actin monomers. This reaction is strongly kinetically disfavored by the presence of actin monomer sequestering proteins within cells. Thus, protein cofactors that promote actin filament nucleation are required to generate actin networks at specific locations within the cell such as at the distal lamellipodium.

One mechanism by which new filaments are nucleated is by creating branches off of the sides of existing filaments via the Arp2/3 complex. This highly conserved seven-protein complex has intrinsically low activity and requires activating protein co-factors. The best studied of these co-factors is the WASP family of nucleation promoting factors (NPFs) that is regulated by Rho-family GTPases¹. Originally, there were only two known activators of the Arp2/3 complex (WASP and SCAR), but that number is grown recently to include several other proteins^{2, 3}. This increasing complexity of Arp2/3 complex activators suggests that cells use subcellular and context specific activation of Arp2/3 for a more robust and precise regulation of branched actin networks.

A more recently discovered mechanism for generating new actin filaments is through a protein called SPIRE⁴, which promotes filament nucleation by bringing monomers together with four tandem actin monomer-binding WH2 domains. The four bound actin monomers are lined up end-to-end and mimic a short single strand of a nascent filament. Together, these monomers form the pointed end of a new filament to which free monomers then bind to grow the nascent filament. SPIRE-mediated nucleation does not result in branched actin filaments, and thus may be utilized either to jump-start network formation or in circumstances in which a stiff branched network is not necessary.

Using protein homology searching, Zuchero et al.⁵ identified the p53 cofactor JMY as having a potential Arp2/3 regulatory sequence. JMY is known to bind to p300/CBP and cooperates

^{*}Correspondence: James E. Bear 450 West Dr., CB#7295 21-227 LCCC Chapel Hill, North Carolina 27599 Telephone: (919) 966-5471 Fax: (919) 966-3015 jbear@email.unc.edu.

with it to activate p53-dependent transcription⁶, but no connection with the actin cytoskeleton had previously been suspected. Zuchero et al. purified JMY and biochemically demonstrated that it activates Arp2/3-induced actin polymerization in a dose dependent fashion. Somewhat surprisingly, they also found that JMY was able to catalyze actin polymerization in the absence of Arp2/3. Further examination revealed that JMY, like SPIRE, was able to nucleate new filament formation through tandem WH2 domains. This is the first instance of these two biochemical activities being united in one protein. By both increasing the speed at which new filaments are formed and by harnessing the amplification of polymerization that occurs after activation of Arp2/3, JMY seems to be capable of inducing very rapid assembly of new actin networks.

Zuchero et al.'s examination of JMY in a cellular context revealed a primarily nuclear localization for JMY in most cell types, as would be expected for a p53 regulator. In primary human neutrophils, however, JMY colocalizes with actin filaments at the leading edge, and is excluded from the nucleus. This localization pattern correlates with motility, as HL60 cells only have JMY in the cytoplasmic compartment when differentiated into highly motile neutrophil-like cells. Furthermore, overexpression and knockdown studies demonstrated that JMY expression promotes the rate of cell migration in wound healing assays. These data are consistent with JMY having a role in controlling actin dynamics in highly motile cells (Fig. 1).

As with any newly discovered protein activity, questions quickly outstrip available answers. One interesting question is whether JMY is playing a role in regulating nuclear actin in addition to its known function as a transcriptional regulator. While not well understood, nuclear actin has been linked to transcription, chromatin structure, and nuclear transport⁷. The characteristics of nuclear actin networks, however, are fundamentally different than those utilized during cell motility, so it is unclear how JMY may function in this context or what consequences any activity would have.

JMY's relationship with other nucleators also remains unclear. There may be cytoplasmic competition for actin monomers, and migration of JMY into that compartment could diminish the supply of actin available for other nucleators such as WASP-Arp2/3 or the formins (another class of actin nucleating factors). Such competition might complicate the analysis of JMY depletion phenotypes due to potential over-activation of other nucleators in JMY's absence. An alternate possibility is that the nucleators act independently, and JMY introduction can be used simply as a context-dependent catalyst. In this fashion, JMY may be part of a network of nucleators and NPFs that act in concert to fine-tune cytoskeletal dynamics.

Another unanswered question concerns the mechanism by which JMY activity is regulated. Given its potent biochemical activity, perhaps it is not surprising that the primary localization of JMY is in the nucleus. Rather than regulating JMY by phosphorylation or other post-translational modification, sequestration of the protein away from the majority of actin could be an ideal way to keep its activity moderated until required. The method of transport from the nucleus and its potential triggers remain to be discovered. Similarly, the supply of JMY protein could be regulated by proteasome-mediated degradation. DNA damage causes an accumulation of JMY protein, while Mdm2-catalyzed ubiquitination targets JMY for proteasome-dependent degradation⁸. Cell motility-related cues could also tap into this mechanism, and contribute to its availability to alter actin dynamics. It will be interesting to determine whether localization and protein degradation are indeed utilized to control JMY activity, and to see whether other mechanisms also contribute.

Another complex issue is reconciling the involvement of JMY in two very different cellular processes: transcription and regulation of actin dynamics. It will be important to examine both

pathways when examining JMY function in the future. Specifically, it will be necessary to test whether any given phenotype is attributable to one or a combination of both activities. Such dichotomy is not unprecedented, as the beta-catenin protein is known to have important roles as a cytoskeletal linker mediating cell adhesion, as well as acting as being a component of the WNT signaling pathway that translocates to the nucleus after pathway activation⁹. Perhaps this system could offer insights in the best path to follow in understanding how JMY balances such discreet functions.

While there are many unanswered questions regarding how JMY functions and is regulated, it is clear that Zuchero et al. have added another entry to the list of important actin regulatory proteins. Their work has therefore helped refine our understanding of actin dynamics and cell motility.

References

1. Chesarone MA, Goode BL. Actin nucleation and elongation factors: mechanisms and interplay. *Curr Opin Cell Biol.* 2009
2. Campellone KG, Webb NJ, Znameroski EA, Welch MD. WHAMM is an Arp2/3 complex activator that binds microtubules and functions in ER to Golgi transport. *Cell* 2008;134:148–161. [PubMed: 18614018]
3. Linardopoulou EV. Human subtelomeric WASH genes encode a new subclass of the WASP family. *PLoS Genet* 2007;3:e237. [PubMed: 18159949]
4. Quinlan ME, Heuser JE, Kerkhoff E, Mullins RD. Drosophila Spire is an actin nucleation factor. *Nature* 2005;433:382–388. [PubMed: 15674283]
5. Zuchero JB, Coutts AS, Quinlan ME, La Thangue NB, Mullins RD. p53-cofactor JMY is a Multifunctional Actin Nucleation factor. *Nature Cell Biology.* 2009This Issue
6. Shikama N. A novel cofactor for p300 that regulates the p53 response. *Mol Cell* 1999;4:365–376. [PubMed: 10518217]
7. Vartiainen MK. Nuclear actin dynamics--from form to function. *FEBS Lett* 2008;582:2033–2040. [PubMed: 18423404]
8. Coutts AS, Boulahbel H, Graham A, La Thangue NB. Mdm2 targets the p53 transcription cofactor JMY for degradation. *EMBO Rep* 2007;8:84–90. [PubMed: 17170761]
9. Perez-Moreno M, Fuchs E. Catenins: keeping cells from getting their signals crossed. *Dev Cell* 2006;11:601–612. [PubMed: 17084354]

Figure 1. JMY Functions both in and out of the nucleus

JMY functions in concert with p300 to activate p53-dependent transcription. In highly motile cells, JMY is transported to the cytoplasm where it promotes the formation of actin filament networks via two separate biochemical activities. It is able to nucleate new filament formation via a SPIRE-like mechanism that is dependent on its tandem actin-monomer binding WH2 domains. In addition, JMY promotes actin branch formation by activating the Arp2/3 complex with a WH2 domain, an actin and Arp2/3 binding central domain (C) and an Arp2/3-binding acidic domain (A). It remains undetermined whether JMY regulates nuclear actin dynamics.