JCB Journal of Cell Biology

SPOTLIGHT

Dendritic actin delivery service

Yun-Jin Pai and Adrian W. Moore

The mechanisms by which the actin cytoskeleton regulates dendritic branching are not fully understood. Nithianandam and Chien (2018. *J. Cell Biol.* https://doi.org/10.1083/jcb.201711136) discover actin blobs, new structures that mediate dynamic actin delivery within a growing dendrite arbor and that mark sites of future branch formation.

As beautifully illustrated in the pioneering drawings of Ramon y Cajal (1995) over a century ago, the neuronal dendrite arbor is an expansive and highly branched cellular structure. To build this arbor is a major cell biological undertaking. A nascent neuron is a small spherical or bipolar cell, and yet it grows an arbor that may have many hundreds of branches spread over an area several hundred-fold larger than that of the original cell (Fig. 1).

Arbor outgrowth and branching require the delivery and organization of large amounts of F-actin. This is a cytoskeleton component used in construction of high-order and terminal branches as well as spines. These branches form from interstitial filopodia that emerge from the main dendrite trunk (Portera-Cailliau et al., 2003). How actin is delivered and positioned for the formation of these new branches has remained unclear.

In this issue, Nithianandam and Chien examine spatial F-actin organization and dynamics in the growing dendrite by live imaging with genetically encoded F-actin probes; they discover a new mechanism that links dynamic actin delivery to the site of future branch formation. They describe spots containing preassembled F-actin termed "blobs" that move bidirectionally through the growing arbor and stall at sites where new branches subsequently emerge. After branch initiation, the blobs further contribute F-actin into the nascent branch (Fig. 1).

How are blobs formed? Nithianandam and Chien (2018) introduced a mutant actin transgene, which creates F-actin that is resistant to severing. They found that this overstabilization of F-actin in dendrites caused the actin blobs to drastically reduce in number but not propagation speed. Subsequently, the number of new branches forming and the growth rate of new as well as existing dendrites were also reduced. These findings were recapitulated in loss-of-function experiments of the actin severing protein Cofilin, which they identified in a genetic screen for regulators of the actin blobs. Actin blob splitting events were also severely reduced by loss of Cofilin or transgenic actin stabilization, suggesting that blob production depends on F-actin severing by Cofilin.

Actin blobs are an important new complement to other actin-organizing systems in the dendrites: actin waves (Ruthel and Banker, 1998) and actin patches (Andersen et al., 2005; Hou et al., 2015). Actin waves start at the base of dendrites and move slowly toward the tip. As the wave front passes, local actin is transiently organized into a growth cone–like series of filopodia and lamellipodia, which is subsequently disassembled at the rear of the wave. In theory, the passing of an actin wave could accelerate local interstitial branch formation, although it remains unclear whether this occurs. Actin patches have a clearer role in branch initiation: they form in situ through local F-actin nucleation at branching sites and then transform into a filopodia. Compared with these previously reported mechanisms, actin blobs are different. In contrast to actin patches, blobs deliver F-actin to sites of branch formation. In contrast to actin waves, blobs can stop and start moving and they go in both directions along the dendrite. Interestingly, some actin blobs moving from retracting dendrites were used to support new branch formation in neighboring areas of the arbor, suggesting they might mediate competition for resources between forming dendrite branches. Both waves and patches also form in axons, along with actin trails that extend from F-actin hotspots on stationary endosomes (Ganguly et al., 2015). Actin trails may be specific to axons, and it is possible that actin blobs are similarly confined to dendrites.

Neurons elaborate arbors with branching morphologies that are characteristic for each neuron type (Ramon y Cajal, 1995). What cell biological mechanisms give rise to this diversity? Previous studies showed that differences in postmitotic transcription factor and F-actin bundling protein activities help differentiate terminal branch morphology between class IV neurons and the closely related class III (Jinushi-Nakao et al., 2007; Nagel et al., 2012). Nithianandam and Chien (2018) find that there is also differential utilization of actin blobs for branch initiation between these two neuron classes. In class IV neurons, the large majority of high order branches form from

Neurodiversity, RIKEN Center for Brain Science, Wako, Saitama, Japan.

Correspondence to Adrian Walton Moore: adrian.moore@riken.jp.

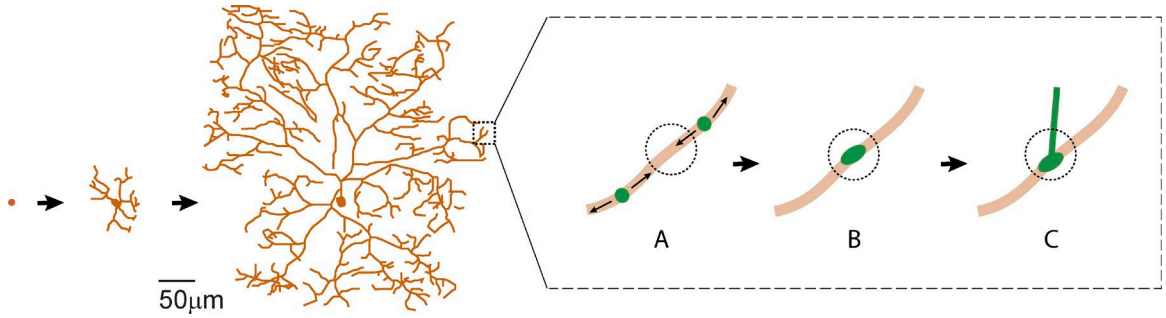

Figure 1. **Arbor outgrowth and branching of a class IV dendrite arborization neuron, beginning from a spherical neuron cell to a complex, branched structure.** Preassembled F-actin blobs (shown in green) exhibit bidirectional movement along the dendrite branch (A) before they stall at a future branch site (B). Finally, they contribute actin to formation of a new branch (C).

the sites where actin blobs stall, but in class III neurons, only a minority do. The authors suggest differential use of blobs as a mechanism for arbor diversification.

Like most important findings, those of Nithianandam and Chien (2018) stimulate more questions. Regarding arbor branch patterning, an earlier study emphasized the importance of actin patch formation to initiate class III terminal branches (Andersen et al., 2005). Nithianandam and Chien (2018) observe that some class IV branches also form from an actin patch (more surprisingly, a small number of branches grow at sites with no evidence of prior F-actin buildup). Therefore, it is worth considering that blobs and patches are convergent processes to initiate branches, and it is subsequent outgrowth programs that create arbor diversification. Moreover, a causal relationship between blobs and branch sites remains unclear; does blob stalling induce a branch site, or do blobs stall at sites already selected for outgrowth? While the internal actin organizations that make up waves, patches, and trails have been probed, it requires further analysis in the blobs. Intriguingly, blobs remain stationary at the branch site for minutes before branch initiation. What, then, is the switch that signals onset of the initiation step, and is it linked to changes in internal actin structure within the blob?

Certainly, with their new findings, Nithianandam and Chien (2018) make an important inroad toward revealing how actin is delivered within the growing arbor and how branch position is selected. These are critical steps through which a neuron constructs one of the most complex and challenging cellular structures—the dendrite arbor.

Acknowledgments

Y.-J. Pai is funded through a RIKEN Special Postdoctoral Researcher Fellowship.

The authors declare no competing financial interests.

References

Andersen, R., Y. Li, M. Resseguie, and J.E. Brenman. 2005. Calcium/calmodulin-dependent protein kinase II alters structural plasticity and cytoskeletal dynamics in Drosophila. *J. Neurosci.* 25:8878–8888. https://doi.org/10.1523/JNEUROSCI.2005-05.2005

Ganguly, A., Y. Tang, L. Wang, K. Ladt, J. Loi, B. Dargent, C. Leterrier, and S. Roy. 2015. A dynamic formin-dependent deep F-actin network in axons. *J. Cell Biol.* 210:401–417. https://doi.org/10.1083/jcb.201506110

Hou, W., M. Izadi, S. Nemitz, N. Haag, M.M. Kessels, and B. Qualmann. 2015. The Actin Nucleator Cobl Is Controlled by Calcium and Calmodulin. *PLoS Biol.* 13:e1002233. https://doi.org/10.1371/journal.pbio.1002233

Jinushi-Nakao, S., R. Arvind, R. Amikura, E. Kinameri, A.W. Liu, and A.W. Moore. 2007. Knot/Collier and cut control different aspects of dendrite cytoskeleton and synergize to define final arbor shape. *Neuron.* 56:963–978. https://doi.org/10.1016/j.neuron.2007.10.031

Nagel, J., C. Delandre, Y. Zhang, F. Förstner, A.W. Moore, and G. Tavosanis. 2012. Fascin controls neuronal class-specific dendrite arbor morphology. *Development.* 139:2999–3009. https://doi.org/10.1242/dev.077800

Nithianandam, V., and C.T. Chien. 2018. Actin blobs prefigure dendrite branching sites. *J. Cell Biol.* https://doi.org/10.1083/jcb.201711136

Portera-Cailliau, C., D.T. Pan, and R. Yuste. 2003. Activity-regulated dynamic behavior of early dendritic protrusions: evidence for different types of dendritic filopodia. *J. Neurosci.* 23:7129–7142. https://doi.org/10.1523/JNEUROSCI.23-18-07129.2003

Ramon y Cajal, S. 1995. Histology of the nervous system of man and vertebrates. Oxford University Press, New York.

Ruthel, G., and G. Banker. 1998. Actin-dependent anterograde movement of growth-cone-like structures along growing hippocampal axons: a novel form of axonal transport? *Cell Motil. Cytoskeleton.* 40:160–173. https://doi.org/10.1002/(SICI)1097-0169(1998)40:2%3C160::AID-CM5%3E3.0.CO;2-J