Developmental Cell
Previews

CellPress

Moving the ER tip by tip

Ha H. Nguyen[1] and Ginny G. Farías[1,*]
[1]Cell Biology, Neurobiology, and Biophysics, Department of Biology, Faculty of Science, Utrecht University, Utrecht 3584 CH, the Netherlands
*Correspondence: g.c.fariasgaldames@uu.nl
https://doi.org/10.1016/j.devcel.2021.12.001

Endoplasmic reticulum (ER) and microtubule (MT) interactions have been observed in different cell types. However, how these interactions are regulated remains unknown. In this issue of *Developmental Cell*, Nourbakhsh et al. show that an ER-localized kinase, TAOK2, catalyzes the dynamic tethering of the ER tip to the MT tip.

For almost four decades, interactions between the tubular network of the endoplasmic reticulum (ER) and microtubules (MTs) have been observed in different cell types. MTs contribute to the formation, elongation, and transport of ER tubules, and in return, ER tubules play a role in MT organization. This indicates a strong interplay between these two tubular structures (Koppers et al., 2020). How this interaction is regulated remains unknown. Tethering proteins play essential roles in the formation and maintenance of contacts between cellular structures. For interactions between organelles and MTs, adaptor proteins that tether the organelle to a specific molecular motor are known to mediate the transport of mitochondria and late endosomes and/or lysosomes (Koppers et al., 2020). Different mechanisms have been reported for the elongation and transport of ER tubules in association with MTs. These are sliding and hitchhiking mechanisms, in which an ER tubule (directly or indirectly) associates with a MT-driven motor to be transported along stable MTs, and the polymerizing and depolymerizing tip attachment complex (TAC) mechanisms pTAC and dTAC, in which the tip of an elongating ER tubule attaches to the growing or shrinking dynamic MT tip (Waterman-Storer and Salmon, 1998; Guo et al., 2018) (Figure 1A). Although motors involved in sliding and hitchhiking mechanisms have been reported, the tethering factors and regulators remain largely unknown. For the pTAC mechanism, the binding of the MT-plus-end protein EB1 to the ER-tip-associated protein STIM1 creates forces that cause membrane deformation and ER extension (Grigoriev et al., 2008). However, the question remains: How is this interaction between the ER tip and MT tip regulated? In this issue, Nourbakhsh et al. (2021) report that an ER-localized kinase regulates the dynamic tethering of the ER tubule tip to the MT tip via its kinase activity (Nourbakhsh et al., 2021).

The authors identified TAOK2 (Thousand and One amino acid Kinase 2), a serine/threonine kinase, as an ER membrane protein that localizes to discrete ER subdomains in human cells. Interestingly, TAOK2 directly associates with MTs via its extreme C-terminal domain, and expression of this domain leads to MT bundling and increased MT acetylation. Simultaneous visualization of the ER membrane, MTs, and TAOK2 revealed that nearly 90% of TAOK2 puncta localize to the tip of ER tubules at contact sites with MTs; this finding suggests that this protein could serve as a tethering factor. To further study the role of TAOK2 as a tethering factor, the authors generated TAOK2 knockout (KO) cell lines. Compared to control cells, in which the ER extends its tubules toward the cell periphery and forms extensive contacts with MTs, TAOK2 KO led to reduction in peripheral ER density and its dissociation from MTs. Loss of TAOK2 also increased ER motility, but not associated with EB3 comets. Motor-dependent transport of ER tubules is in fact more frequently observed and faster than TAC-mediated transport (Guo et al.,2018, Grigoriev et al.,2008), which suggests that motor-based mechanisms could compensate. However, because ER density in the cell periphery was reduced, it remains unclear if this is the case. In TAOK2 KO cells, EB3 velocity also increased, while EB3 comet tracks became less directional and more frequently paused; these findings suggest a role for TAOK2 in maintaining MT dynamics. TAC-dependent ER movement was previously shown to be mediated by STIM1 and EB1/3 (Grigoriev et al., 2008). In this study, TAOK2 KO led to a drastic reduction in STIM1 accumulation at MT plus-ends and disrupted TAC-mediated ER movements. Immunostaining revealed that TAOK2 colocalized with more than 30% of EB1 comets and TAOK2 co-immunoprecipitated with EB1 and STIM1. These results suggest that TAOK2 acts as a regulator of STIM1-EB1 interaction (Figure 1A). At endogenous levels, from all EB1 associated with the ER, 62.5% of these contacts were positive for TAOK2 puncta. However, endogenous STIM1 and TAOK2 puncta displayed just a 20% of co-distribution at ER tips. In addition, Nourbakhsh et al., found in the C-terminal domain of TAOK2 a highly conserved EB-binding motif, Ser-x-Ile-Pro (SxIP), which is also present in STIM1 and other EB-binding proteins. It remains unknown whether TAOK2 can promote ER elongation independent of STIM1, as TAOK2 could directly bind EB through its SxIP domain.

The importance of TAOK2 in TAC-mediated ER-MT interaction prompted the authors to investigate what regulates this interaction and whether the kinase activity of TAOK2 is involved. Thus, they used a TAOK2 kinase-dead mutation (K57A) and the TAOK2 kinase inhibitor CP43, and they found that the ER-MT tethering activity is negatively regulated by TAOK2 kinase activity (Figure 1B). Loss of TAOK2 catalytic function results in stronger tethering between the ER and MTs, leading to reduced ER motility and MT growth. This kinase activity also results in differences in the ER-MT tethering required in interphase and mitotic cells. During metaphase, when chromosomes are aligned by mitotic spindles, the ER and TAOK2 both reorganize at each end of the spindle poles but are largely excluded from chromosomes and regions in between the two poles. TAOK2 KO cells showed a defect

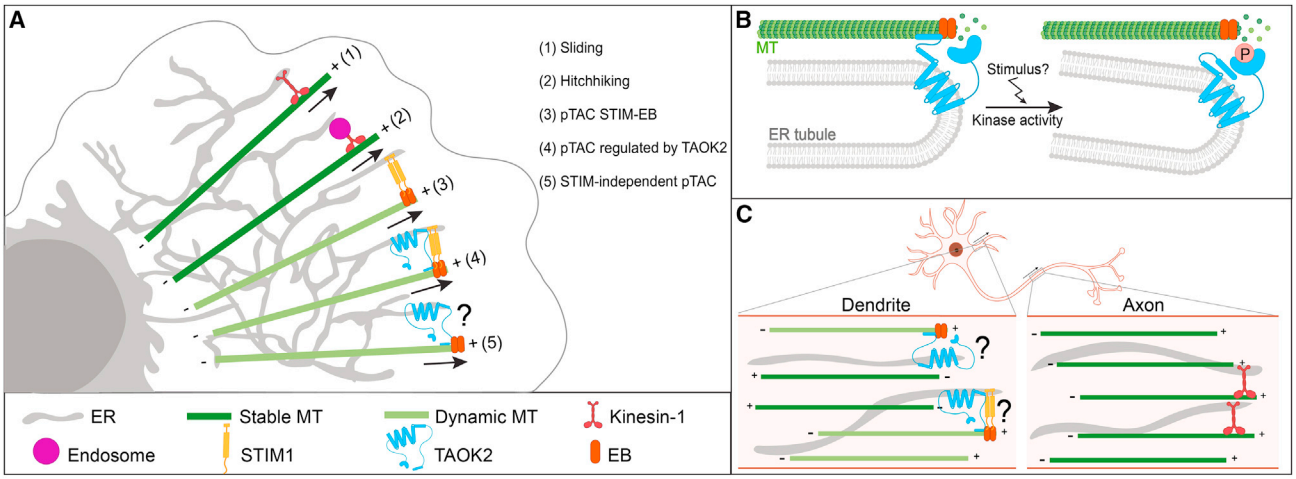

Figure 1. TAOK2, a recently identified ER-MT tethering factor, regulates MT-plus-end-directed ER transport
(A) Mechanisms involved in MT-based transport of ER tubules to the cell periphery.
(B) ER-MT contact is regulated by TAOK2 kinase activity.
(C) Different mechanisms involved in the transport of ER tubules into the axon and dendrites. Potential role of TAOK2 in TAC-mediated ER transport into dendrites.

in ER membrane morphology and a reduction in ER association with spindle poles. Moreover, the kinase-dead TAOK2 caused the opposite effect, a failure of disengagement of the ER from mitotic spindle MTs. Both caused severe mitotic defects which were observed as impaired bipolar spindle formation and chromosome misalignment (Nourbakhsh et al., 2021). These results indicate an important role of TAOK2 kinase autoregulation for proper ER-MT reorganization during cell growth.

The study from Nourbakhsh et al. (2021) has identified a crucial player in ER-MT tethering and dynamics. Their findings raise several new questions. What is the function of TAOK2 in differentiated and highly polarized cells such as neurons? TAOK2 has previously been shown to be involved in dendritic development, and it was linked to neurodevelopmental disorders (Nourbakhsh and Yadav, 2021). However, little is known about its molecular function in neurons. Important ER _reorganization occurs during neuronal development, in which ER tubules need to be transported to the distal tips of the axon and dendrites. ER tubule transport into the axon is mediated by the MT-driven motor kinesin-1, whereas its transport into dendrites appears to involve a TAC-mediated mechanism (Farías et al., 2019) (Figure 1C). Could TAOK2 be involved in this TAC-mediated ER tubule extension into dendrites? What signals could regulate the activity of TAOK2? Could TAOK2 catalytic domain phosphorylate essential players in the ER-MT _interface to regulate their interactions? Understanding how TAOK2 responds to different signals and potentially impacts the dynamic ER-MT tethering could provide new insights into the importance of ER-MT communication.

Different mechanisms mediate ER elongation along MTs. What is the relevance of having multiple ways to pull ER tubules into the cell periphery? Is there crosstalk and regulation between sliding mechanisms (direct or hitchhiking) and TAC? Are these different mechanisms required for efficient transport of a particular set of ER tubules with specific functions, or are they simply redundant mechanisms? Previous *in vitro* studies have suggested a cooperation of sliding and TAC movements in membrane tubule extension along MTs (Rodríguez-García et al.,2020). Further research into the dynamics and interdependency of different ER extension mechanisms could give us a better overview of how ER-MT interplay is maintained and regulated during cell growth and differentiation under physiological and pathological conditions.

DECLARATION OF INTERESTS

The authors declare no competing interests.

REFERENCES

Farías, G.G., Fréal, A., Tortosa, E., Stucchi, R., Pan, X., Portegies, S., Will, L., Altelaar, M., and Hoogenraad, C.C. (2019). Feedback-Driven Mechanisms between Microtubules and the Endoplasmic Reticulum Instruct Neuronal Polarity. Neuron *102*, 184–201.e8.

Grigoriev, I., Gouveia, S.M., van der Vaart, B., Demmers, J., Smyth, J.T., Honnappa, S., Splinter, D., Steinmetz, M.O., Putney, J.W., Jr., Hoogenraad, C.C., and Akhmanova, A. (2008). STIM1 is a MT-plus-end-tracking protein involved in remodeling of the ER. Curr. Biol. *18*, 177–182.

Guo, Y., Li, D., Zhang, S., Yang, Y., Liu, J.J., Wang, X., Liu, C., Milkie, D.E., Moore, R.P., Tulu, U.S., et al. (2018). Visualizing Intracellular Organelle and Cytoskeletal Interactions at Nanoscale Resolution on Millisecond Timescales. Cell *175*, 1430–1442.e17.

Koppers, M., Özkan, N., and Farías, G.G. (2020). Complex Interactions Between Membrane-Bound Organelles, Biomolecular Condensates and the Cytoskeleton. Front. Cell Dev. Biol. *8*, 618733.

Nourbakhsh, K., and Yadav, S. (2021). Kinase signaling in dendritic development. Front. Cell. Neurosci. *15*, 624648.

Nourbakhsh, K., Ferreccio, A.A., Bernard, M.J., and Yadav, S. (2021). TAOK2 is an ER-localized kinase that catalyzes the dynamic tethering of ER to microtubules. Dev. Cell *56*.

Rodríguez-García, R., Volkov, V.A., Chen, C.Y., Katrukha, E.A., Olieric, N., Aher, A., Grigoriev, I., López, M.P., Steinmetz, M.O., Kapitein, L.C., et al. (2020). Mechanisms of Motor-Independent Membrane Remodeling Driven by Dynamic Microtubules. Curr. Biol. *30*, 972–987.e12.

Waterman-Storer, C.M., and Salmon, E.D. (1998). Endoplasmic reticulum membrane tubules are distributed by microtubules in living cells using three distinct mechanisms. Curr. Biol. *8*, 798–806.