

DEEP LEARNING-BASED ANALYSIS OF OPTICAL NANOSCOPY IMAGES REVEALS ACTIVITY-DEPENDENT REORGANIZATION OF THE PERIODICAL ACTIN LATTICE IN DENDRITES

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The discovery of the actin-spectrin membrane-associated lattice using super-resolution optical microscopy changed our understanding of the organization of actin, a key protein of the neuronal cytoskeleton. This lattice, containing actin rings periodically spaced 180nm apart, was initially discovered in axons [1] and later observed in dendrites of multiple types of neurons [2,3]. However, its role and regulation mechanisms remain unknown. We demonstrate that neuronal activity regulates the actin-based lattice in dendrites but not in axons of cultured rat hippocampal neurons. Using STimulated Emission Depletion (STED) nanoscopy, we observed complex and diverse patterns of fluorescently-labelled F-actin inside neuronal processes. While the F-actin periodical ring patterns were robustly detectable in axons, dendrites exhibited patches of actin rings perpendicular to the shaft, mixed with patches of longitudinal fibers parallel to the shaft axis. This diversity of patterns posed a colossal challenge for quantitative analysis, which we addressed using a deep learning segmentation approach. We trained a fully convolutional neural network to identify the periodical actin lattice and longitudinal actin fibers in STED images of fixed neurons. With this approach we could quantify the extent of the periodical lattice and the fibers on a large dataset and at various neuronal activity levels. We observed that increasing neuronal activity leads to the reorganization of the periodical lattice in dendrites but that it remains stable in axons.

REFERENCES

- [1] Xu, Ke, Guisheng Zhong, and Xiaowei Zhuang. "Actin, spectrin, and associated proteins form a periodic cytoskeletal structure in axons." *Science*, **339**, 452-456, (2013).
- [2] Zhong, Guisheng, et al. "Developmental mechanism of the periodic membrane skeleton in axons." *Elife*, **3**, e04581, (2014).
- [3] D'Este, Elisa, et al. "STED nanoscopy reveals the ubiquity of subcortical cytoskeleton periodicity in living neurons." *Cell reports*, **10**, 1246-1251, (2015).