**Adaptive Force Generation by Active Motors and Passive Crosslinkers in Microtubule Overlaps**

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Conventionally, the mechanical forces driving the sliding of overlapping microtubules relative to each other have been attributed to the action of molecular motors and the dynamics of cytoskeletal filaments, which both consume chemical energy. By contrast, non-enzymatic filament cross-linkers have been regarded as mere friction-generating entities. Recently, we experimentally demonstrated that diffusible microtubule crosslinkers of the Ase1/PRC1/Map65 family can also generate directed microtubule sliding when confined between partially overlapping microtubules [1]. We quantitatively explain the underlying force generation by the entropic expansion of confined Ase1 molecules diffusing within the microtubule overlaps. The thermal motion of passive crosslinkers is thus harnessed to generate mechanical work analogous to compressed gas propelling a piston in a cylinder. Strikingly, the Ase1-generated forces were sufficient to antagonize the sliding of overlapping microtubules driven by kinesin-14, Ncd, a motor which is diffusively anchored to one microtubule via its tail domain while translocating another microtubule by its non-processive motor domain [2]. Currently, we are extending our studies towards motor proteins, which are capable of combining active force generation and entropic expansion in one kind of molecule. For example, we found that the sliding velocity of microtubules driven by human kinesin-14, HSET, decreases when microtubules start to slide apart, resulting in the maintenance of finite-length microtubule overlaps [3]. We quantitatively explain this feedback by the local interaction kinetics of HSET with overlapping microtubules causing retention of HSET in the shortening overlaps. Consequently, the increased HSET density in the overlaps leads to a density-dependent slowdown of the sliding velocity and the generation of an entropic force antagonizing the force exerted by the motor domains. Our results demonstrate that the spatial arrangement of microtubules can regulate the collective action of molecular motors through local alteration of their individual interaction kinetics.

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[2] A. Lüdecke, A. M. Seidel, M. Braun, Z. Lansky, S. Diez, Diffusive tail anchorage determines velocity and force produced by kinesin-14 between crosslinked microtubules, under review (2017).

[3] M. Braun, Z. Lansky, A. Szuba, F. W. Schwarz, A. Mitra, M. Gao, A. Lüdecke, P. R. ten Wolde, S. Diez, Changes in microtubule overlap length regulate kinesin-14-driven microtubule sliding, DOI: 10.1038/NCHEMBIO.2495 (2017, in press).