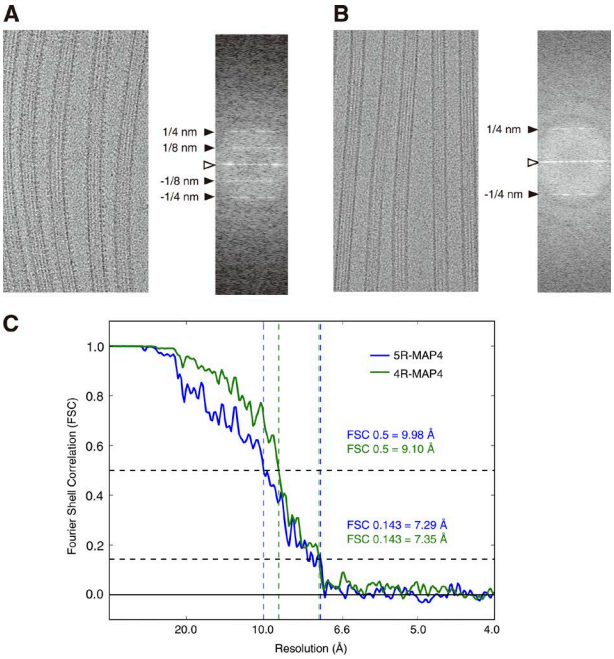
Supplemental material

Shigematsu et al., https://doi.org/10.1083/jcb.201711182



D

Data collection statistics

	5R-MAP4	4R-MAP4
Microscope	Tecnai Arctica	Tecnai Arctica
Voltage (kV)	200	200
VPP	no	no
Camera	Falcon II	Falcon II
Defocus range (μm)	1.5 - 2.5	1.5 - 2.5
Exposure time (s)	2.0	2.0
Dose rate (e⁻/pixel/s)	45	45
Frames	34	34
Total Dose (e⁻/Å²)	55	55
Nominal pixel size	1.284	1.284
Number of particles	15,148	77,616
Resolution (overall)	7.29	7.35

Microtubule lattice statistics

	5R-MAP4	4R-MAP4
Helical rise (Å)	8.7	8.7
Helical twist (degrees)	-25.76	-25.76
Axial rise (Å)	81.26	81.44
Axial twist (degrees)	-0.46	-0.45

Figure S1. **Resolution and statistics for the cryo-EM reconstructions; related to Fig. 2. (A)** Example micrograph of 4R-MAP4–kinesin-1–microtubule complex (left). Fast Fourier transform of the micrograph is also shown (right). **(B)** Example micrograph of 4R-MAP4–microtubule complex (left). Fast Fourier transform of the micrograph is also shown (right). **(C)** Fourier shell correlation curves of the MAP4–kinesin-1–microtubule complexes. **(D)** Data collection and processing statistics and helical parameters for the reconstructions. VPP, Volta phase plate.

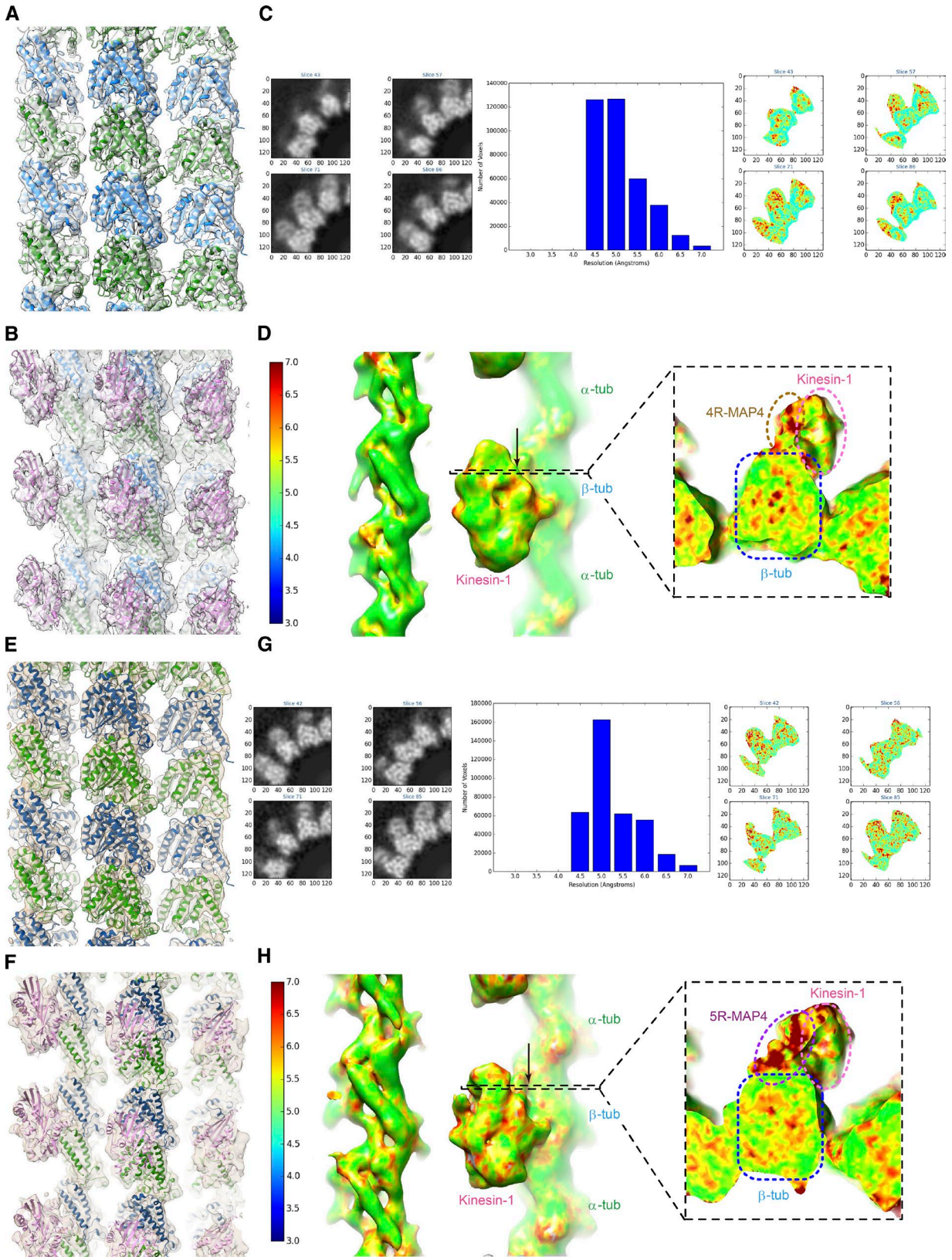


Figure S2. **Local resolution estimates; related to Fig. 2. (A and B)** The density maps and the docked atomic models of the 4R-MAP4–kinesin-1–microtubule complex. **(C and D)** Local resolution estimates of 4R-MAP4–kinesin-1–microtubule complex calculated by ResMap. Inset in D shows the horizontal slice observed from the plus end indicated by the arrow in the middle panel. **(E and F)** The density maps and the docked atomic models of the 5R-MAP4–kinesin-1–microtubule complex. **(G and H)** Local resolution estimates of 5R-MAP4–kinesin-1–microtubule complex calculated by ResMap. Inset in H shows the horizontal slice observed from the plus end indicated by the arrow in the middle panel.

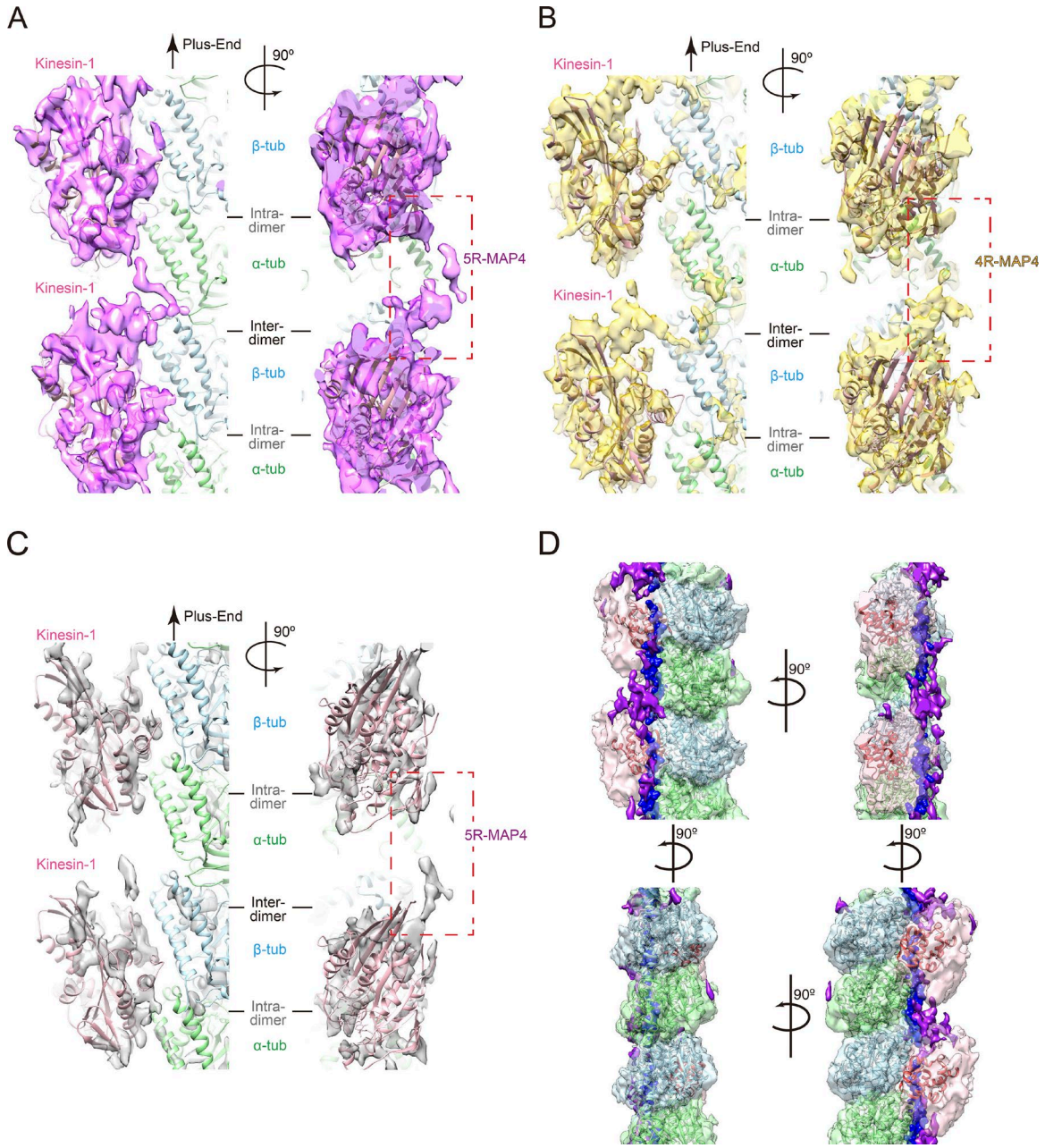


Figure S3. **Dynein does not compete at the microtubule binding site with Tau family MAPs; related to Fig. 3. (A–C)** Difference maps between 5R-MAP4–kinesin-1–microtubule complex and kinesin-1–microtubule complex (A), between 4R-MAP4–kinesin-1–microtubule complex and kinesin-1–microtubule complex (B), and between 5R-MAP4–kinesin-1–microtubule complex and 4R-MAP4–kinesin-1–microtubule complex (C) generated by UCSF Chimera. The occupancy and/or stability of kinesin-1 were different among these three maps so that the densities corresponding with MAP4 as well as kinesin-1 appeared. **(D)** Cryo-EM reconstruction of 5R-MAP4–kinesin-1–microtubule complex shown with the microtubule binding domain (MTBD) of dynein (red ribbon) and Tau (blue surface). Green, α-tubulin; light blue, β-tubulin; pink, kinesin-1; purple, 5R-MAP4. In contrast with kinesin-1, MTBD of dynein does not collide with 5R-MAP4 or Tau.