

Supplementary Materials:

Structures of actin-like ParM filaments show architecture of plasmid-segregating spindles

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Video 1. 4.3 Å cryo-EM reconstruction of ParM+AMPPNP filaments.

The cryo-EM structure of the ParM+AMPPNP filaments is shown as an isosurface contoured 2σ away from the mean. A small interface holds the two protofilaments together. Five ParM monomers that were refined into the density are shown.

Video 2. Morph between the ParM+AMPPNP filament and the ParM+ADP filament structures.

A morph was generated between the PDB coordinates of the ParM+AMPPNP and the ParM+ADP filament structure at the filament level (top) and at the monomer level (bottom). Only main chain atoms are shown in the morph as ribbon diagrams. ParM+AMPPNP is shown in green while ParM+ADP is shown in blue. All intermediate morph states are shown at a lower brightness level because they do not represent observed data, and are merely shown for illustrative purposes.

Video 3. ParM doublets do not show any super-helical twist.

Sequential z-slices of a reconstructed electron cryotomogram of ParM doublets *in vitro*. These tomography data show clearly the absence of any super-helical twist in the filaments.

Video 4. ParM doublet model.

Model of the ParM antiparallel doublet is shown. The volumes have been filtered down to 40 Å to better illustrate how the two filaments in the doublet are out of phase with each other.

Video 5. ParM doublets are observed in *E. coli* cells containing plasmids with the ParMRC locus.

A video containing sequential z-slices of a reconstructed electron cryotomogram of an *E. coli* cell transformed with the low-copy number plasmid (pKG491) ('mini-R1' replicon). A ParM doublet is highlighted.

Video 6. ParM bundles and doublets observed in *E. coli* cells containing plasmids with the ParMRC locus.

Video containing sequential z-slices of a reconstructed electron cryotomogram of an *E. coli* cell transformed with the medium-copy number plasmid (pKG321). Both ParM doublets and bundles were observed in this cell.