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The tension mounts at centromeric loops

Study reveals how pericentric chromatin generates tension between sister centromeres.

Centromeres are more than just simple stretches of DNA that recruit the kinetochore proteins required to bind spindle microtubules and segregate chromosomes during mitosis. In fact, the chromatin surrounding centromeres is organized into a spring-like structure that bridges the centromeres of sister chromatids and resists the pull of spindle microtubules. The tension generated when sister kinetochores are correctly attached to microtubules emanating from opposite spindle poles helps silence the mitotic checkpoint and allow sister chromatid segregation. Lawrimore et al. now describe how the pericentric chromatin spring establishes intracentromere tension (1).

Kerry Bloom and colleagues at the University of North Carolina at Chapel Hill previously demonstrated that condensin and cohesin proteins accumulate on the pericentric chromatin of budding yeast chromosomes and organize it into a "bottle brush" configuration, with numerous DNA loops radiating out from a central axis that runs between sister kinetochores in parallel to the spindle axis (2, 3). Now, Bloom says, his team wanted to understand how this structure helps to generate tension between sister centromeres.

To learn more about the forces produced at centromeres, Bloom and colleagues, led by graduate student Josh Lawrimore, fluorescently labeled a segment of pericentric chromatin and followed its movements in metaphase yeast (1). Surprisingly, Lawrimore et al. found that pericentric chromatin moved in similar ways regardless of whether or not cells were treated with benomyl, an inhibitor of microtubule dynamics. During metaphase, therefore, most centromeric chromatin movements are not driven by microtubules. "That means there's a force regime near to the centromere that's largely microtubule independent," Bloom explains. Instead, the researchers determined, the movements of pericentric chromatin are mainly driven by a combination of Brownian motion and the random, ATP-dependent activities of chaperones and chromatin-remodeling proteins.

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To characterize the pericentric forces produced by these microtubule-independent movements, Lawrimore et al. used a special plasmid that carries two centromeres separated by a relatively short stretch of DNA. Almost all of this intervening, pericentric DNA can be fluorescently labeled, allowing the researchers to assess its overall structure when the plasmid was aligned on the metaphase spindle. The researchers found that, even in the presence of benomyl, the plasmid's pericentric chromatin was stretched out along the spindle axis, so much so that some of its nucleosomes would have to be unwrapped and released in order for it to extend so far.

Thus, thermal fluctuations and random, ATP-dependent enzyme activities generate a force that stretches pericentric chromatin outwards along the spindle axis. Polymer physics provides a simple explanation for this (4, 5). As they fluctuate, the pericentric chromatin loops will tend to collide and repel each other, creating an outward-directed force that can counteract the natural tendency of chromatin—or any linear polymer—to curl up into a random coil. "So thermodynamic principles tell you that pericentric chromatin is going to be under tension," Bloom says. By organizing pericentric chromatin into a bottle brush configuration, condensin and cohesin generate sufficient tension to separate sister centromeres, even without the input of spindle microtubules. "We view this as a sort of primordial segregation machine that predisposes sister centromeres to sit on the surface of chromosomes opposite from one another," Bloom explains.

Moreover, Bloom and colleagues think that, by dynamically altering the number of DNA loops emanating from its central axis, pericentric chromatin can act as a "shock absorber" to buffer the variable forces produced by spindle microtubules as they stochastically grow and shrink. Accordingly, Lawrimore et al. found that changes in intracentromere tension can affect the dynamics of kinetochore-attached microtubules. The researchers now want to learn more about how condensin and cohesin are recruited to pericentric chromatin and how they generate its bottle brush structure.

1. Lawrimore, J.G., et al. 2015. *J. Cell Biol.* http://dx.doi.org/10.1083/jcb.201502046
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3. Stephens, A.D., et al. 2013. *J. Cell Biol.* 200:757–772.
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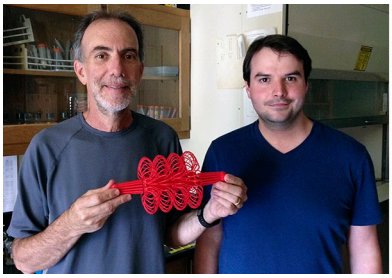
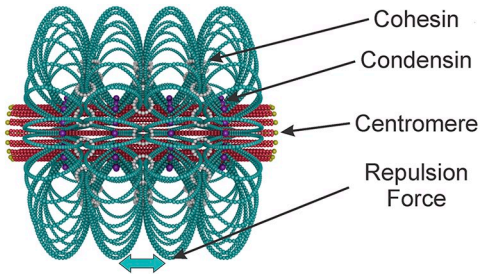
Focal Point





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Kerry Bloom (left) and Josh Lawrimore (right) hold a 3D-printed model of the yeast mitotic spindle. A 2D version (right) highlights the "bottle brush" structure of pericentric chromatin, in which condensin (purple) and cohesin (grey) organize the DNA surrounding centromeres (yellow) into radial loops (teal) emanating from a central axis (red) that lies parallel to the spindle axis. Lawrimore et al. reveal that repulsion between the radial loops generates tension between sister centromeres independently of the mitotic spindle. The structure of pericentric chromatin may also allow it to act as a shock absorber that buffers the variable forces generated by dynamic spindle microtubules.