Text by Mitch Leslie
mitchleslie@comcast.com

Passenger proteins check in

Despite their name, the passenger proteins aren't just along for the ride during mitosis. They are busy helping control the attachment of spindle fibers to the chromosomes and ensuring that the cell splits after the chromosomes part.

INCENPs' many locations

But it was the proteins' seemingly bizarre movements that first caught the eye of William Earnshaw (now at the University of Edinburgh in the UK) and his colleagues in 1987. Previous work had demonstrated that sister chromatids can separate even when researchers cut the microtubules that form the spindle, suggesting that the centromere might house motors that propel the chromosomes. But cell biologists knew little about the centromere's architecture. In 1985, Earnshaw's lab snared the first three of the structure's components, which they dubbed the centromere proteins, or CENPs (Earnshaw and Rothfield, 1985). Using antibodies against the chromosomes' protein scaffold, Carol Cooke then identified a pair of proteins that cluster on the centromere. The Edinburgh team called this pair the inner centromere proteins, or INCENPs (Cooke et al., 1987).

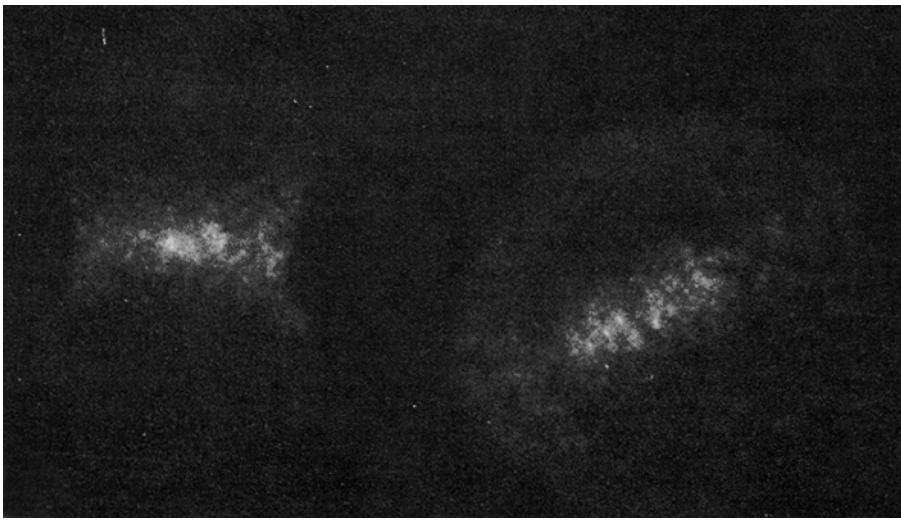
Carol Cooke and William Earnshaw identify the first passenger proteins and catalogue their strange movements.

Earnshaw and colleagues could see INCENPs clinging to the arms and centromere in mitotic chromosomes. They took a closer look, treating cells so that the chromosomes were spread-eagled into the textbook "X" shape. They saw that some INCENPs adhered to the centromeres internal to the CENPs at the last points of contact between sister chromatids.

Biochemical tests indicated that the INCENPs clung tightly to the chromosome's protein scaffold, so the researchers got a jolt when they tracked the proteins through mitosis. At the beginning of anaphase the INCENPs appeared to jump ship. Instead of following the chromosomes as they slid apart to the poles, the proteins festooned the microtubules in the middle of the mitotic spindle. And some snuggled up to the cell membrane at the point where the cleavage furrow later squeezes the cell in two. "Nobody had seen a chromosomal protein change its position that dramatically," says Earnshaw. He and his colleagues had identified the first passenger protein (the two INCENPs turned out to be splice variants of the same molecule).

Many places, many functions

INCENP concentrated at the site of presumptive cleavage furrow formation before myosin appeared there (Eckley et al., 1997), making it tempting to speculate that it was being dropped at the site after hitching a ride on the chromosomes as a "passenger." But earlier functions were still under consideration. Earnshaw had found that cells injected with anticentromere antibodies failed to align chromosomes during metaphase and, although they kept going through mitosis, they did so with extremely defective spindles (Bernat et al., 1990). "This led me to think that the chromosomes must normally 'give' something to the spindle in metaphase that helped stabilize it in anaphase," he says. The candidate for that something was INCENP.



EARNSHAW

INCENPs cluster at the middle of each cell even at late- (left) or mid-anaphase (right).

Later work showed that INCENP partners with three other passenger proteins—Aurora-B, survivin, and borealin—to form the chromosomal passenger kinase complex. This complex targets many proteins in the cell including histone H3, which acquires phosphate tags as the chromatin condenses at the onset of mitosis. The passenger complex also helps to fasten microtubules to the centromeres and to choreograph the separation of the two daughter cells. Just last year, the labs of Earnshaw and Hironori Funabiki (Rockefeller University, New York, NY) uncovered borealin (Gassmann, 2004), also known as Dasra (Sampath et al., 2004), and showed that it's crucial for correctly attaching microtubules to the centromeres, stabilizing the spindle, and completing cell division. When Earnshaw and colleagues watched INCENP flitting about in 1987, they were seeing the protein complex on the job. JCB

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