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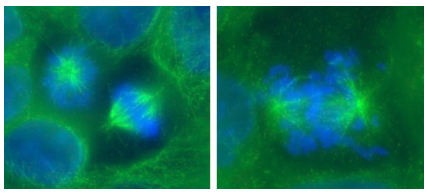
Demystifying Aurora B

In 2003, two papers uncovered the mitotic functions of Aurora B using novel small molecule inhibitors.



ADAPTED FROM DITCHFIELD ET AL., 2003

Compared with control cells (left), inhibition of Aurora B (right) causes impaired alignment of chromosomes (blue) on the mitotic spindle (green) at metaphase.

During mitosis and meiosis, the spindle assembly checkpoint (SAC) acts to ensure that anaphase is only initiated once all chromosomes achieve attachment to both spindle poles. In the early 2000s, scientists studying mitosis in budding yeast found that the kinase Ipl1 activates the SAC in response to loss of tension at kinetochores (1) and also destabilizes inappropriately attached kinetochores (2). An Ipl1 orthologue in higher organisms, Aurora B, had known roles in several mitotic processes and was under intense study. But its function was poorly understood until the back-to-back publication of a pair of papers in *JCB* in 2003 (3, 4).

Curiously, neither set of authors—Stephen Taylor's group at the University of Manchester in the UK, and Jan-Michael Peters's lab at the Institute of Molecular Pathology in Vienna, Austria—initially planned to venture into the competitive Aurora B field. Taylor had just returned to Manchester after completing his postdoctoral studies on mitosis when he gave a seminar to former colleagues at the pharmaceutical company AstraZeneca. "Afterward, Nick Keen approached me, saying he had a new compound he thought might interest me," recalls Taylor.

The drug had been developed as a possible inhibitor of the Aurora family protein Aurora A. Taylor and postdoc Claire Ditchfield agreed to have a look.

Meanwhile, Peters's lab was about to begin characterizing a novel, polyploidy-inducing kinase inhibitor developed by Boehringer Ingelheim. Peters's group wanted to test whether Boehringer's compound targeted separase, a protein required for the separation of sister chromatids early in anaphase. "Luckily, Silke Hauf, a postdoc in my lab, was willing to embark on this high-risk project. We had no idea whether we could identify the target of this compound or how interesting it would be," says Peters.

"I wasn't thinking about risk at that time. I just thought it sounded like a fun project. But in retrospect I would completely agree. It was a bit risky," laughs Hauf.

Unbeknownst to one other, Hauf and Ditchfield each took a "chemical genetics" approach, using an array of assays to study their drug's effects on mitotic phenotypes. Both their compounds caused defects in chromosome alignment and segregation. Despite this, cells still entered anaphase—pointing to impaired SAC—then failed cytokinesis and reentered mitosis.

Crucially, the scientists also quickly discovered that their compounds blocked phosphorylation of histone H3 at serine 10—which had recently been demonstrated to be a target of Aurora B (5). "Then the question became, what is it that Aurora B is doing to allow proper chromosome segregation?" says Peters.

Peters's group noticed that Aurora B inhibition hampered cells' ability to recognize and correct syntelic attachments, which result when both of a chromosome's kinetochores become attached to the same spindle pole. A compromised SAC allowed anaphase onset in spite of these misattachments, thereby causing aneuploidy.

> "What is it that Aurora B is doing to allow proper chromosome segregation?"

"Where it got really interesting for us," says Taylor, "was when we saw that our compound potently overrode the SAC in the presence of taxol, but not as efficiently in the presence of nocodazole."

That's because taxol and nocodazole each trigger SAC by different mechanisms. Microtubule destabilization by nocodazole prevents any microtubule–kinetochore attachments from forming; the new data showed that unattached kinetochores caused arrest without Aurora B's help, though Aurora B inhibition did help cells exit arrest after a few hours. By contrast, taxol stabilizes microtubules, promoting both correct and incorrect (syntelic) kinetochore attachments. In this case, Aurora B inhibition let cells ignore attachment errors and go straight to anaphase. Observes Peters, "Our data and the data from Taylor's lab suggested that Aurora B senses and then corrects wrong attachments."

Both labs concluded Aurora B likely recognizes syntelic attachments due to insufficient kinetochore tension. Aurora B then evicts misattached microtubules and indirectly triggers SAC arrest by generating unattached kinetochores. But the researchers noted that Aurora B also has a direct role in SAC, because its inhibition blocked kinetochore recruitment of SAC components such as BubR1. This explains why Aurora B inhibition sped exit from nocodazole arrest.

These findings were especially interesting in light of a heated debate then ongoing in the field, which centered on whether the SAC detects the presence of unattached kinetochores, or if it is instead triggered by the lack of physical tension at unattached kinetochores (6). Thus, when Taylor and Peters finally learned of each other's work at a conference in England in 2002, they decided to copublish their results.

"These two papers were significant not only because they identified Aurora B as a central component required for proper kinetochore attachment to the spindle, but also because they illustrated the power of small molecule inhibitors to directly define this function," remarks *JCB* editorial board member Rebecca Heald.

1. Biggins, S., and A.W. Murray. 2001. *Genes Dev.* 15:3118–3129.
2. Tanaka, T.U., et al. 2002. *Cell.* 108:317–329.
3. Ditchfield, C., et al. 2003. *J. Cell Biol.* 161:267–280.
4. Hauf, S., et al. 2003. *J. Cell Biol.* 161:281–294.
5. Hsu, J.Y., et al. 2000. *Cell.* 102:279–291.
6. Maresca, T.J., and E.D. Salmon. 2010. *J. Cell Sci.* 123:825–835.