A gene that silences the X chromosome

Counting to two doesn't sound like an impressive feat, but it's a matter of life and death for women's cells.

The genetic activity of a single X chromosome is sufficient for a human cell, but women must somehow deal with a double dose. Fortunately, mammals have evolved a way to silence one of the two X chromosomes in female cells.

More than 30 years after researchers discovered this phenomenon, however, the mechanisms by which cells count their X chromosomes and inactivate unneeded ones remain elusive.

"People have really scratched their heads over this," says Alan Ashworth of the Institute of Cancer Research in London.

Two new reports add to a growing body of evidence that an unusual gene called *Xist* may hold the key to X chromosome inactivation. While most genes encode proteins, *Xist* produces a short strand of RNA, a nucleic acid similar to DNA.

Several clues point to *Xist*'s importance in silencing one copy of a female's X chromosome. *Xist* is active on the inactivated X chromosome but inactive on the active copy.

Furthermore, the RNA strands made by *Xist* cover the inactivated X chromosome "like a sheath," says Rudolf Jaenisch of the Whitehead Institute for Biomedical Research in Cambridge, Mass.

Jaenisch and other scientists have also shown that mutations in *Xist* prevent inactivation of the X chromosome on which it resides. In recent experiments, he and his coworkers added copies of a small X chromosome fragment that includes *Xist* to another chromosome of a male mouse embryo cell.

This technique tricks the male cell, which apparently calculates that it contains more than one X chromosome, into activating the *Xist* gene and turning off a nearby gene that the researchers had added along with *Xist*, Jaenisch's group reported in the July 12, 1996 CELL.

Jaenisch and Whitehead colleague Jeannie T. Lee now report in the March 20 NATURE that the X chromosome material they add seems to have the ability to shut down an entire chromosome.

When the pair examined the activity of four genes spread along chromosome 12, to whose tip they had added the X chromosome fragments, they found all four genes silent. The researchers also observed that the RNA strands made by the *Xist* genes were spread along the entire length of the chromosome.

The X chromosome fragment added by Jaenisch and Lee is big enough to hold several genes, leaving unsettled the question of whether *Xist* acts alone.

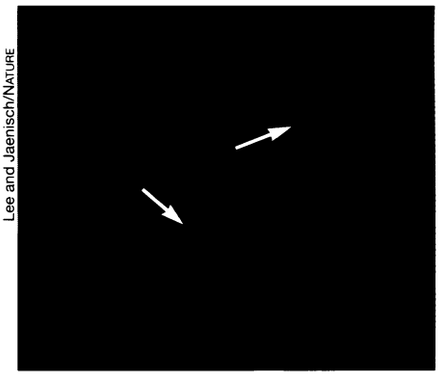
Ashworth and his colleagues have started to address that issue by working with a smaller piece of the X chromosome, containing *Xist* and little else. Like Jaenisch's group, Ashworth's team has added this genetic material to non-X chromosomes in male mouse cells.

Once again, the male cells seem to think they contain more than a single X chromosome. They activate the added *Xist* genes and sometimes even the *Xist* gene on their one X chromosome.

This result, also published in the March 20 NATURE, strongly implies that cells tally their X chromosomes by simply counting *Xist* genes, says Ashworth.

While this new research has begun to reveal the secrets behind X chromosome inactivation, scientists must still explain how a cell counts *Xist* genes and how the *Xist* RNA silences the thousands of genes on an X chromosome.

Answering the latter question may help explain how other animals manage their sex-related chromosomal differences. In fruit flies, for example, females do not turn off an X chromosome, and males hyperactivate their single copy so that it does the job of two.



Lee and Jaenisch/NATURE

Arrows point to the chromosome 12 regions (light blue) where copies of the Xist *gene were added. The genes produce strands of RNA (red) that spread along the chromosome and somehow inactivate other genes.*

While that action seems the opposite of inactivating a chromosome, some recent research hints that fruit flies, like mammals, use RNA to change the X chromosome's activity, observes geneticist Helen K. Salz of Case Western Reserve University School of Medicine in Cleveland. —*J. Travis*

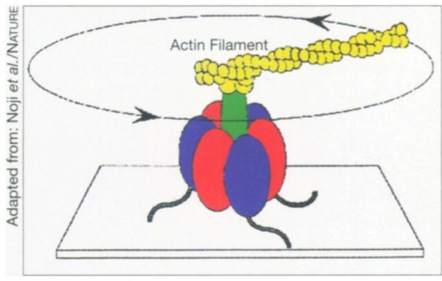
Molecular motor spins out energy for cells

With parts that resemble pistons and a drive shaft, the enzyme F_1-ATPase looks suspiciously like a tiny engine. Indeed, a new study demonstrates, that's exactly what it is. A movie of a single enzyme molecule in action shows that it spins like a motor to crank out ATP, the ubiquitous molecule that provides energy for biochemical processes in cells.

F_1-ATPase is a subunit of a larger enzyme, ATP synthase, that spans the membranes of mitochondria, the energy-producing organelles in cells. Scientists from the Tokyo Institute of Technology and Keio University in Yokohama anchored molecules of F_1-ATPase to a glass slide and—like putting a flag on top of a pole—attached a long, fluorescent filament of actin to the end of the drive shaft. By bathing the enzyme in ATP, the researchers made F_1-ATPase break down the energy molecule and watched as it whirled the fluorescent filament around like a propeller.

"I think this is going to be a classic contribution to the field," says Paul D. Boyer of the University of California, Los Angeles. Boyer proposed almost 20 years ago, before F_1-ATPase's complete structure was known, that it must rotate to catalyze ATP formation. This enzyme is the first one known to operate by this mechanism, he says.

Previously, the smallest molecular motors scientists had studied were those that drive bacterial flagella. F_1-ATPase is less than one-tenth that size. The Japanese team plans to make some "moving toys" by inserting the mitochondrial enzyme between a bacterial flagellum



Adapted from: Noji *et al.*/NATURE

The enzyme F_1-ATPase, attached to a glass slide, spins a fluorescent actin filament counterclockwise.

and a plastic bead, says the Tokyo Institute's Masasuke Yoshida. The group's findings appear in the March 20 NATURE.

Researchers previously had indirect biochemical and spectroscopic evidence that the enzyme rotates, an idea also supported by F_1-ATPase's three-dimensional structure, determined in 1994. However, conflicting results left some scientists skeptical. The current study should erase those remaining doubts, says Richard L. Cross of the State University of New York Health Science Center in Syracuse. "It's so visual that it's easy for everyone to understand."

The enzyme puts out a very large torque, considering that the actin filament is more than 100 times the length of the enzyme itself, Yoshida says. "Can a man rotate a 150-meter rod?" The enzyme can spin a long filament because it ratchets down the rotation rate when it carries a heavy load, he explains, suggesting that F_1-ATPase can change gears—as a good motor should. —*C. Wu*