CELL BIOLOGY

Chromosome Yield New Clue To Pairing in Meiosis

For the organisms that indulge in sexual reproduction, the essence of the act is pairing—male with female, sperm with egg. But one pairing step comes even earlier than that. During the first phase of meiosis, the specialized form of cell division that produces the sperm and eggs, the chromosomes themselves pair up, with each chromosome from an individual's father joining with its counterpart from the mother before the developing gamete divides and the chromosomes are partitioned into the two daughter cells. Exactly how the chromosome partners find each other, pair up, and then segregate into the daughters has been a mystery, but in this issue of *Science*, a research team from the Salk Institute provides a clue.

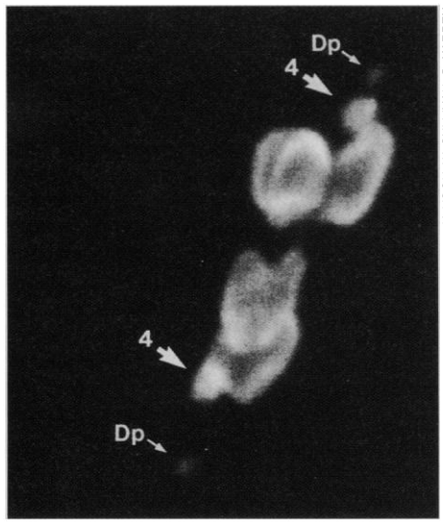
On page 118, Gary Karpen and his Salk colleagues Mong-Huong Le and Hiep Le report that at least some of the chromosome pairings that take place during meiosis in the fruit fly *Drosophila melanogaster* are initiated or maintained by the heterochromatin, a type of DNA found in the chromosomes' central and terminal regions that consists mainly of noncoding repetitive sequences. Work that will soon be published by another group, including Abby Dernburg of Stanford University, John Sedat of the University of California, San Francisco, and Scott Hawley of UC Davis, reportedly comes to a similar conclusion.

Terry Hassold, a mammalian meiosis researcher at Case Western Reserve University in Cleveland, says the two groups have produced "elegant studies that define a role for a previously enigmatic part of the eukaryotic genome." He was referring to the heterochromatin, sometimes considered to be "junk DNA" because of its lack of genes, even though it makes up about a third of the genome in flies and up to 25% in humans.

And beyond that, researchers hope that the new results will not only provide a clearer picture of normal chromosome pairing and segregation during meiosis, collectively called "disjunction," but also help them understand what causes this process to go awry. When that happens, gametes that either lack an entire chromosome or carry an extra copy are produced. In humans, these errors can result in conditions such as Down syndrome.

Both teams are looking at a relatively simple variant of meiotic disjunction. For most chromosomes to segregate normally during meiosis, they must first exchange genetic material when they pair up, in a process called recombination. But in the female fruit fly, chromosome 4, the smallest chromosome, always fails to recombine with its partner before separating from it, and other chromosomes occasionally skip the recombination step. For these nonrecombining chromosomes to pair and segregate, another system, called achiasmate disjunction, has to come into play.

But even though achiasmate disjunction was discovered some 60 years ago, its molecular mechanism has remained unclear. For nearly 30 years, the dominant hypothesis was one proposed in the 1960s by Rhoda Grell of Oak Ridge National Laboratory in Tennessee. She concluded that size similarity is what enables homologous chromosomes to find each other and pair up during achiasmate disjunction. Then about 4 years ago, cytological and genetic evidence suggested to Hawley's team that heterochromatin structure plays a key role. But the chromosomes studied by Grell and Hawley were too big and too complex to characterize molecularly, and thus it was hard to tell who was right.



K. YOOK, W. SULLIVAN, G. KARPEN

Moving apart. In this micrograph of segregating fruit fly chromosomes, the arrows point to the minichromosome (Dp) and chromosome 4.

And that is where the work of Karpen and his colleagues comes in. "What he's done is make the mechanism of chromosome segregation tangible at the molecular level," says meiosis expert Terry Orr-Weaver of the Whitehead Institute in Cambridge, Massachusetts. This was possible because the researchers began with a fruit fly "minichromosome," created decades ago in lab fruit flies, that undergoes achiasmate disjunction. Because the minichromosome contains only 1300 kilobases of DNA, its molecular composition, which consists mostly of heterochromatin, could be pinned down.

To determine which chromosomal regions are needed for the minichromosomes to pair up during meiosis, the Salk workers first generated a series of mutant minichromosomes from which specific regions were deleted. They then introduced these mutated minichromosomes in pairs of varying composition into fruit flies, which were subsequently mated to see how the minichromosomes segregated during the formation of the flies' germ cells.

The group could trace the fate of the minichromosomes because they also carried marker genes, for eye or body color, that enabled the researchers to spot which progeny produced by the matings inherited which minichromosomes. By counting flies that inherited each color characteristic, the researchers could tell whether disjunction had been normal, with each fly inheriting one minichromosome, or whether nondisjunction had occurred, with some flies inheriting two minichromosomes while others got none.

The results point to the importance of the minichromosomes' heterochromatin for normal achiasmate disjunction. The key factor, Karpen and his colleagues found, was the extent of overlap in the heterochromatin sequences on the pairing minichromosomes. "The degree of heterochromatin overlap is crucial," Karpen says. "If the chromosomes overlap by 1000 kilobases, they do well." But as the amount of overlap decreased below that, the frequency of nondisjunction increased linearly.

The Karpen team's evidence also suggests that the heterochromatin comes into play during chromosome pairing, rather than the segregation that follows. They found, for example, that giving flies extra copies of the *nod* gene, which encodes a protein that helps keep achiasmate chromosomes attached to the meiotic spindle, did not improve disjunction rates for any minichromosome pairing. Because the NOD protein acts after chromosome pairing, this result implies that the problems with the mutant minichromosomes occurred in that earlier step.

Further evidence that heterochromatin is responsible for joining rather than segregating the chromosomes comes, Karpen says, from Dernburg, Sedat, and Hawley, who have a paper in press in *Cell*.* This work, which Karpen cites (with the authors' permission) in his *Science* paper, used fluorescent microscopy to show that the paired chromosomes are held together by their heterochromatin. "The Hawley paper demonstrates very nicely by a very different

* Hoping to include the authors' description of their work, *Science* offered to delay this News report until 12 July, *Cell*'s publication date. But *Cell* Editor Benjamin Lewin instructed the authors not to discuss their work with *Science* before 12 July.

method that heterochromatic regions on homologous chromosomes are associated with one another," Karpen says.

Other researchers familiar with the work, which both teams have presented at meetings, say the two sets of results complement each other very well. "I think it's important that there are two halves to this story," says Orr-Weaver. "Karpen sees the end product of the meiotic event, while Hawley looks at an intermediate stage."

But even though heterochromatin has now been firmly implicated in chromosome pairing for achiasmate disjunction, many questions remain. One is just how the heterochromatin draws the chromosomes together. It may do so directly, if similar sequences on the two chromosome partners pair up, or indirectly, via proteins that bind to the heterochromatin DNA on both chromosomes and then to each other. Also unclear is whether heterochromatin plays a role in normal disjunction, in which recombination occurs.

And then there is the big question of whether the *Drosophila* results will reveal anything about meiosis in higher animals, including humans. No one knows whether chromosomes in these organisms undergo achiasmate disjunction, says Case Western's Hassold. Still, he notes, it is "not unreasonable" to expect that they might. He points out, for example, that some of the chromosomal abnormalities seen in *Drosophila nod* mutants, which disrupt achiasmate disjunction, resemble the chromosomal abnormalities that can arise when human meiosis goes awry. If achiasmate disjunction does occur in higher animals, then fruit flies, like birds and bees, may have plenty to say about the basics of human reproduction.

–Jean Marx

OPTICS

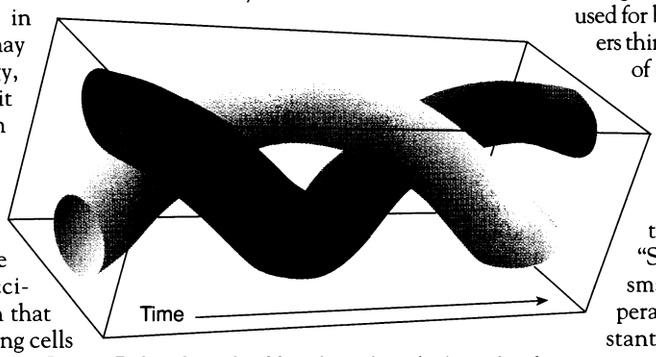
Helical Beams Give Particles a Whirl

For the past 25 years, physicists wanting to pick up and hold particles as small as a single molecule have reached for the tweezers—the optical tweezers, that is. Consisting of finely focused laser beams, optical tweezers rely on the electric fields created by light to trap particles that are too small to manipulate mechanically. Now two teams of researchers, in the United Kingdom and in Australia, have found that by using specially sculpted laser beams they can do more than just hold a particle in place: They can make it spin, speed up and slow down its spinning, and even bring it to a stop and spin it the other way. Such dexterity, say the researchers, makes the device an entirely new tool—what the British group calls an optical spanner, or wrench.

Described this week at the International Quantum Electronics Conference in Sydney, Australia, the spanner may one day be useful in nanotechnology, say researchers who envision using it to build tiny machines or power them by rotating particular parts. What's more, the spanner can immobilize its targets with less intense beams than optical tweezers use. Less intensity means less heating of the target, and—through a fortuitous accident—the British team has shown that the spanner may be able to hold living cells without killing them. If so, says biologist Justin Molloy of Britain's University of York, the spanner's gentler touch "could enable new classes of experiments to be performed using monomeric proteins."

Optical tweezers use the electric field created at the narrowest point of a tightly focused laser beam to hold onto particles of dielectric, or insulating, materials—which include most biological samples. The particles are electrically attracted to the region where the field, and the beam, is strongest: right at the center.

But Miles Paget and his colleagues at St. Andrew's University in Scotland and, working independently, Halina Rubinsztein-Dunlop's group at the University of Queensland in Australia suspected that a laser beam with a different intensity profile, known as Laguerre-Gaussian, might create a more versatile kind of trap. Such laser beams are doughnut-shaped in cross section, with a dark spot in the middle surrounded by a bright ring of laser power, and are created by shaping the beam with cylindrical lenses or holograms. Like an optical tweezers, a Laguerre-Gaussian beam draws a specimen toward its most intense regions—which are found not at its center but in the bright ring. If the size of the specimen is roughly the same as the ring diameter, it gets pinned by its edges. Both groups managed to trap particles this way.



Doing the twist. Map shows how the intensity of a Laguerre-Gaussian beam changes with time.

SOURCE: *PHYSICAL REVIEW LETTERS*, VOL. 75, NO. 5, 31 JULY 1995

But that is not all that Laguerre-Gaussian beams can do. In their simplest form, such beams have a spinning, helical electromagnetic field. St. Andrew's physicist Les Allen predicted in 1992 that these rotating beams could impart angular momentum to a trapped particle, making it spin. Both groups have now shown that Allen was correct. In a paper soon to appear in *Physical Review A*, the Queensland researchers describe how, by changing the optics and reversing the orientation of the Laguerre-Gaussian mode, they were able to speed up and slow down a trapped, rotating particle and reverse its direction.

Both teams also found that their technique required less beam power than a conventional optical tweezers setup does to immobilize a particle—just one third of the usual power, the St. Andrew's group reports in a paper that will soon be published in the *Journal of Modern Optics*. This has important implications for biological samples, some of which are "opticuted" by the beam of conventional optical tweezers. Although they have yet to do a structured experiment, Neil Simpson of the St. Andrew's team says that they carried out an accidental one when one of their samples became contaminated with living cells. The researchers are not sure exactly what kind of cell ended up trapped in their beam, but it seems to have survived the experience. Although optical tweezers have already been used for biological experiments, the researchers think the spanner could extend the reach of these optical tools in this field.

Some biologists are not convinced that this promise will be realized, however. Molecular biologist Steven Block of Princeton University in New Jersey believes the beam will still damage samples. "Significant absorption of light by a small living particle leads to a temperature increase which may be too substantial to be consistent with biological experimentation," he says.

Proponents of nanotechnology have fewer reservations about the new tool. Peter Houzego of PA Consulting Group, a British technology R&D company, says it will find plenty of uses "when we start getting down to nanotechnology [and] we're going to need to be manipulating materials almost at the molecular level." When that day comes, engineers everywhere may want an optical spanner in their toolbox.

–Sunny Bains

Sunny Bains is a writer in Edinburgh, U.K.