Do Chromosomes Cross Talk?

If chromosomes were organized in a constant pattern in the nucleus they might be able to converse with their neighbors

"Anything that gets us away from thinking about the nucleus in terms of a loose assortment of molecules is to be encouraged," says Eric Davidson, a molecular biologist at the California Institute of Technology, commenting on indications from a number of sources that chromosomes are arranged in a predictable and ordered fashion in the nucleus. "The evidence looks very impressive," he adds, referring particularly to Michael Bennett's work at the Plant Breeding Institute, Cambridge, England.

The cytology literature from back into the 19th century is peppered with hints of order in the nucleus, but with no clear demonstration of the fact, still less with any rationale of how it might come about. Researchers in several laboratories in the United States are now producing data that are persuasive of consistently ordered nuclei. Bennett's work not only offers such evidence but also adds a rationale for the laws underlying order.

Cytologists' persistent search for signs of organization in the nucleus has been motivated largely by the need to understand the behavior of chromosomes at certain times of the cell cycle but particularly during the formation of gametes. When sex cells are formed during meiosis, homologous chromosomes must "find" each other prior to segregation of the complete diploid complement into two equivalent haploid sets. Could the pairing and subsequent separation of homologs be a case of order coming out of chaos, or is there an underlying pattern that aids the attainment of that order at meiosis?

While he was trying to answer this question Bennett came across some observations of perhaps even greater fundamental importance. "If chromosomes really are arranged in a consistent pattern," suggests Bennett, "then it is possible that the genes on the arm of one chromosome might affect the activity of genes on the arm of an adjacent chromosome if they are usually neighbors."

"There is no direct evidence in support of this proposal," says Richard Flavell, a geneticist at the Plant Breeding Institute, "but it is provocatively intriguing." Gabriel Dover, of the Department of Genetics at the University of Cambridge, remarks that "There are so many surprises at what you find going on in the eukaryotic genome these days, so this idea might well turn out to be true, but there is no way of knowing at the moment." If it is true, adds Dover, "it would be extremely important." John Sedat, of the University of California, San Francisco, is sure it is true. "There's a lot of old data to support it," he insists.

Nature and scientific technique have, however, conspired to hinder the search for order in the nucleus. For a start, individual chromosomes undergo subtle structural changes during the cell cycle that make them virtually indistinguishable under the microscope for much of the time. Added to this, most of the methods cytologists have developed for looking at chromosomes involve first squashing the nucleus, so that any order that there might have been inevitably is largely destroyed.

The early conviction that chromosomes are arranged according to some regular rules continued until the 1930's and then evaporated. Terry Ashley, of the University of Tennessee, and N. Pocock, of Calgary, Canada, therefore felt able to make the following observation earlier this year: "Despite the extensive evidence to the contrary, it has been widely assumed for many years that chromosomes are randomly arranged in the nucleus. Starting with this basic assumption, the means by which homologs 'find' one another at meiosis becomes one of the most perplexing problems faced by cytogeneticists today."

Ashley's own work, however, points to some kind of association between chromosomes in the nucleus, and this, she infers, might reflect a higher level of organization. "I chose to work with the lily (*Ornithogalum virens*) because it has only three pairs of chromosomes," she explains. "You therefore have a fair chance of seeing what order exists." Ashley has found that in haploid cells the three chromosomes are joined to each other at their tips, the telomeres, in a specific order. She also notes that the telomeres are attached to the surface of the inner nuclear membrane.

Montrose Moses, of Duke University, has noted similar association between chromosome ends and nuclear membrane. He says he favors the notion of specific attraction between pairs of telomeres that brings the chromosomes together, rather than a physical tethering of pairs by, for instance, microfibrils. Whatever mechanism might be operating in bringing the ends of chromosomes together, it is clear that the observation of such associations is a strong lead in the search for nonrandomness in the nucleus.

Ashley and Pocock say that such evidence suggests "a degree of order in nuclei that was previously little suspected." Ashley perceived that the length of the chromosome arms was important in the way the chain of chromosomes assembled itself, but she did not arrive at any rules that the process might seem to follow.

Meanwhile, Bennett was attacking the problem from a slightly different direction. He had been trying for 10 years to understand how homologous pairs of chromosomes came together systematically at meiosis. "I was forced to ask myself, how would I design the process if I were an engineer?" he recalls. "I certainly wouldn't start from a random assortment of chromosomes." He began contemplating the most logical ways in which a bag of chromosomes might be arranged so that assembly of homologous pairs could be achieved most efficiently and with least chance of their becoming hopelessly entangled.

The first question to settle was whether, in a diploid cell, the two parental sets of chromosomes, the haploid genomes, are intermingled or are separate from each other. A key element in tackling this was the development of a technique for mapping the positions of the chromosomes within the nucleus. Working with cereal plants, Bennett and his colleagues did this by making sections through target cells that are dividing. By this technique very good images of the chromosomes themselves are displayed, and the centromeres (the constriction in the chromosomes where fibers attach during spindle formation) and the nucleolar organizer region are readily visible. Each chromosome is identified by its characteristic morphology, and a three-dimensional map of the disposition of the centromeres in the cells directly indicates the positions of the chromosomes.

The answer to this first question, as

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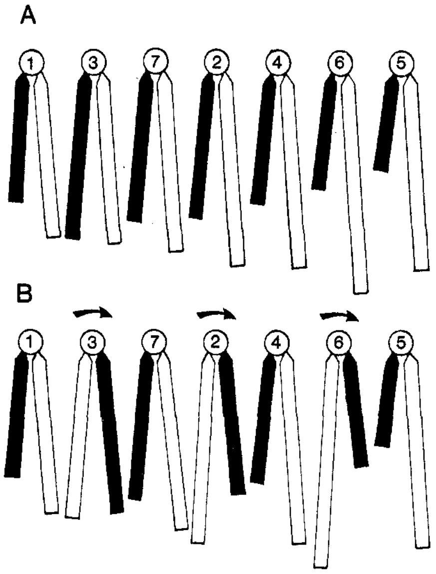
revealed by the thin-sectioning technique, was that the chromosomes in a diploid cell tend to remain segregated as haploid sets. "With this established," says Bennett, "we then had to ask, what is the basis of the haploid genome separation, or to put it another way, how does each haploid set assemble itself?" A crucial lead to the answer came from a paper that had been published almost a decade earlier.

A colleague of Bennett's visited Russia in 1978 and while he was at Novosibirsk he picked up a paper that had been published in 1969 by A. I. Shchapova. When Bennett's colleague returned to the United States, he left the paper, which was in Russian, at the Cambridge Laboratory. "This proved to be an important catalyst," says Bennett.

Shchapova was convinced that chromosomes were highly ordered in the nuclei and she suggested that they were arranged in each haploid set according to the length of the arms. Specifically, she said that the longest arm was joined to the shortest, the next longest to the next shortest, and so on. Such an arrangement would give a virtually constant sum of arm lengths for each pair, thus producing an important regularity.

Shchapova had based her assumptions on data from many species of plants and animals. Bennett tested the idea on just one species, rye, and it looked promising. But there was a problem. "It didn't seem to me that the long-to-short arm connections would work mechanically," says Bennett. "But it did occur to me that if I were to rotate alternate chromosomes in Shchapova's model through 180° and then reinsert them, this would bring together the most similarly sized pairs throughout the haploid complement." This arrangement gives the same chromosome order as that in Shchapova's model and it would work mechanically in real nuclei.

Working with rye and barley, Bennett calculated the 360 possible orders for the seven chromosomes in the haploid complements of sectioned cells. He then ranked these orders according to their increasing separation of centromeres. When he compared the actual order with the prediction, he found a remarkable constancy very close to the optimum. "The model really does appear to work," says Bennett, "and I feel justified in asserting that the disposition of chromosomes is normally highly ordered." Moses describes the work as "an ingenious and convincing study and I don't feel able to knock any holes in it."



Chromosomes in order

The seven chromosomes of rye are arranged as in Shchapova's model (A), that is, with links between the longest arm and the shortest arm, the second longest to the second shortest, and so on. In Bennett's model (B) the order is the same but alternate chromosomes are turned through 180°, thus giving an arrangement that would be mechanically viable in the nucleus.

If the description of chromosome disposition is correct, then the problem of homologous pairing is immediately solved. At its most simplistic, one can imagine the two haploid sets ordered in the same way and facing each other across the nucleus. Pairing would simply be a matter of bringing the sets together, just like pairing one's fingers by closing the hands palm to palm.

The question of how chromosome arms recognize their correct partner remains to be solved of course. But the concept has several interesting implications. For instance, even closely related species can differ in the amount of DNA they have in their nuclei, but this need not necessarily mean that the chromosome order is severely disrupted between the two if the extra DNA is distributed equally over most of the chromosome arms. There are, for instance, two species of salamander, *Plethodon cinereus* and *P. vehicculum*, the second of which has almost twice as much DNA as the first, and yet the ratio of sizes of chromosomes within the species is almost identical. The converse is that a small addition of DNA to a single chromosome could dramatically alter overall disposition, and this might be instrumental in some evolutionary changes.

A second implication is the one already mentioned, concerning cross talk between adjacent chromosomes. Although it now seems clear that chromosomes are arranged in a predictable and ordered manner at some point of the cell cycle, it remains to be demonstrated that this order is maintained throughout the major business period of the cell, the interphase. "I can see no good reason why this should not be so," proposes Bennett, "in which case cross talk must be considered as a possibility."

John Sedat, whose computer simulation work based on chromosomes from *Drosophila* salivary glands makes him a keen proponent of cross talk, pushes the idea even further. "It could be extremely important," he says. "Just as the three-dimensional structure of a protein is what is really important rather than its amino acid sequence, so too could the three-dimensional context of a gene be what matters most." He even suggests that the spatial disposition of chromosomes might alter in a controlled way during development, "and this could affect the activity of genes at different times during development, according to which other genes they are brought near to."

All this might seem farfetched, and there is a good deal of healthy skepticism about it. But there is an intriguing attraction about it too. "Molecular biology is still trying to grapple with things at the level of gene expression," says Dover. "It's just not possible to deal with this in three dimensions, but one can think of lots of fascinating possibilities."

Members of gene families and other repeated sequences converse with each other, says Dover, especially as they lie close to each other. The effect, often, is for the sequences to be made more or less uniform throughout the genome. Clearly, genes that consistently find themselves as neighbors on adjacent chromosomes would more readily be able to engage in this homogenization process.

And what of jumping genes? More and more it appears likely that genes and other genetic elements may be mobile within the genome. If a copy of a gene jumps out of an established cluster, is it more likely to reinsert itself in a part of the genome in close proximity rather than far away? If close, then adjacent chromosome arms may reflect their long association through sharing many genes and other genetic elements, even though the sequences may have diverged substantially, and might therefore be difficult to recognize immediately as being related.

As Dover commented: almost anything is possible in the eukaryotic genome.—ROGER LEWIN