Unraveling the Genetics of Fragile X Syndrome

Geneticists have been puzzled by fragile X syndrome ever since it was discovered back in the 1970s. From the curious constitution of the affected chromosome to the unusual inheritance pattern of its symptoms, nothing about the syndrome, which is the most common inherited form of mental retardation, has seemed straightforward.

Now, two groups have independently discovered the site of the fragile X mutation. What they are finding is not only helping to explain the peculiar features of the disease, but has also shattered one of the geneticists' most cherished assumptions. "We assume that an inherited mutation is stable," says Grant Sutherland at the Adelaide Children's Hospital in Australia, the leader of one of the groups. "And until now, that has been reasonable." But in results described on pages 1097 and 1179 Sutherland and his colleagues and a team led by Jean-Louis Mandel of INSERM and CNRS in Strasbourg, France, show that the fragile X mutation is itself highly mutable. "To my knowledge there is nothing else like this in any other organism or on any other gene," says Sutherland.

The discovery of the fragile X site should improve prenatal diagnosis of the syndrome, as well as help identify asymptomatic carriers who might pass the defect to their children. Currently, fragile X syndrome can be diagnosed only by looking for a characteristic abnormality in which the tip of the long arm of the X chromosome seems to be connected to the rest of the chromosome by a slender thread. Such chromosomes are easily broken—hence the name fragile X. This gross chromosomal change is rarely evident, however, in asymptomatic carriers of the fragile X defect. But the new work provides the basis for designing molecular probes that can be used to identify such carriers.

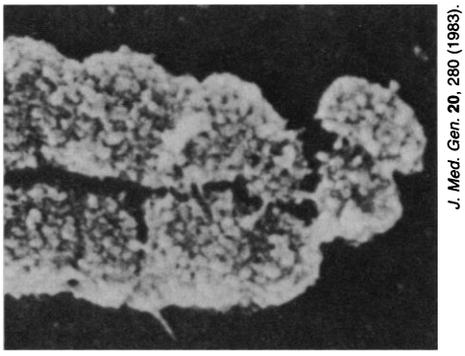
The new finding does not settle all the issues concerning fragile X, however. Indeed, it fuels an ongoing debate over the cause of an inheritance pattern that violates the laws for X-linked diseases. Ordinarily all males who inherit a defect on the X chromosome are affected because they have only one copy of the chromosome, whereas females have a second copy that usually compensates for the defect. But between 20% and 50% of males having the fragile X mutation are asymptomatic. Fragile X genetics is "heresy to a human geneticist," says biologist Charles Laird of the University of Washington at Seattle.

The asymptomatic male carriers of fragile X can pass the gene along to their daughters, who are also asymptomatic, in keeping with the usual inheritance pattern of X-linked diseases. But the pattern takes an unexpected twist again in the third generation: The children of those daughters—the females as well as the males—may have the syndrome. "Something apparently has to happen to the mutation in a female before the deleterious effects are seen in progeny," says Laird.

The new results suggest that at least part of the "something" may be a change in size of the fragile X region. Both the Sutherland and the Mandel groups have found that a DNA fragment from the fragile-X site is much larger in affected individuals than in normal individuals and in asymptomatic male carriers. The fragment size remains virtually unchanged when the male carriers transmit it to their daughters. But in the affected children of those daughters, the fragile-X sites show an astonishing size increase, the researchers found, sometimes containing more than twenty times as much genetic material as in the asymptomatic mothers. "This is not an obligate change," notes Mandel. "But when we see it, it is always when the mutation is passed by a female to her children."

The controversy concerns whether the size change alone suffices to explain the curious emergence of fragile-X symptoms after two generations of asymptomatic carriers. Laird doesn't think so. He suggests that the underlying cause of the inheritance patterns is a phenomenon called genomic imprinting in which genes inherited from the mother behave differently from those inherited from the father as a result of having undergone different chemical modifications in the two parents. And there are signs that imprinting occurs at the fragile X site.

One chemical modification that occurs during imprinting is addition of methyl groups to the DNA, and research in several labs has shown that the fragile X sites of affected individuals contain more methyl groups than those of either normal individuals or asymptomatic carriers. According to Laird, the increased methylation during imprinting is the important factor in determining who actually shows fragile X symptoms. Imprinting in the mother, he suggests, inactivates genes in the fragile X area. As a result, those genes are not expressed in offspring who inherit a female-imprinted fragile X chromosome, a problem that is particularly acute for male offspring, although fragile X symptoms may manifest in female children as well.



J. Med. Gen. **20**, 280 (1983).

Hanging by a thread. *The fragile X chromosome defect is but one of the syndrome's mysteries.*

In Laird's view, the size changes seen by the Sutherland and Mandel groups may be just secondary to the methylation changes during imprinting. Mandel agrees that imprinting is a factor in determining who gets fragile X symptoms, but says that the issue of which comes first—the changes in methylation or the changes in size—has not yet been resolved.

In contrast, Sutherland sees no role at all for imprinting. "Certainly the change in size of chromosomes is adequate for explaining all of fragile X genetics," he says. The fragile X-site can sometimes vary in size from cell to cell in a single individual, he notes, which means that the size variations can potentially occur whenever a cell replicates its DNA and divides—without the DNA having to pass through a mother and acquire an imprint.

In theory, knowing the fragile-X site may help resolve the issue. Laird's hypothesis predicts that the distribution of asymptomatic carrier males to those with symptoms should be equal. This was not previously testable because of the inability to detect the fragile X defect in all asymptomatic carriers. Now, however, researchers can almost always detect even obscure fragile-X sites by a direct DNA analysis of the region. But in a story with as many twists and turns as this one, no one knows quite what to expect. ■ MICHELLE HOFFMAN