THE HUMAN GENE MAP NEAR THE FRAGILE X

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This thesis is submitted as the complete requirement for the degree of Doctor of Philosophy within the Department of Paediatrics, Faculty of Medicine, at The University of Adelaide, South Australia.

August 1990.

First of Two Volumes.
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The fragile X syndrome is the most common cause of familial mental retardation. It is characterised by mental retardation, subtle dysmorphic features, and a fragile site at Xq27.3. Segregation studies have demonstrated incomplete penetrance in males and females.

It was not known whether males with X-linked mental retardation who were fragile X negative had a disorder that was allelic to the fragile X syndrome. The locus responsible for non-specific X-linked mental retardation in a large pedigree was mapped to Xp11 by linkage analysis. This locus (MRX1) is distant from the fragile X locus (FRAXA) which is located at Xq27.3. The fact that MRX1 is not allelic to FRAXA indicates that non-specific X-linked mental retardation is genetically heterogeneous. The 98% confidence interval for MRX1 location was estimated by using a novel resampling strategy to be 0 to 9 centiMorgan (cM) distal to DXS14.

The development of a precise genetic map near FRAXA has been hampered by a lack of closely linked polymorphic loci. The closest restriction fragment length polymorphisms (RFLPs) to FRAXA lay 5 cM proximal (DXS269) and 3 cM distal (DXS304) to FRAXA. The established order of loci near FRAXA was cen-F9-DXS105-DXS98-DXS369-FRAXA-DXS304-DXS374-DXS52-qter.
A panel of 14 cell lines with X chromosome translocation or deletion breakpoints near FRAZA was assembled. The locations of the breakpoints were defined with 14 established probes. Seven of the cell lines had breakpoints between DXS369 and DXS304, and it was not possible to define further the locations of the X chromosome breakpoints in relation to FRAZA. One of these cell lines was derived from a female with Hunter syndrome (MPS II; iduronate-2-sulfatase [IDS] deficiency) due to an X-autosome translocation, thus localizing IDS to between DXS369 and DXS304.

The panel of cell lines was used to localize 18 new DNA probes in this region. One probe was an IDS cDNA clone; the remainder were anonymous DNA fragments. Eight of the probes detected loci near FRAZA. The X chromosome breakpoints, the new probes, and IDS were all localized in relation to each other and to FRAZA. The order of probes and loci near FRAZA was:


RFLPs were detected by the probes VK21 and VK23. RFLPs were also detected at IDS using the IDS cDNA clone. RFLPs were not detected with the probes VK16 or VK18. The RFLPs were mapped in normal pedigrees using the LINKAGE package of computer programs and programs written by the candidate. The following order of loci and recombination fractions were obtained:
This genetic map was used as the basis for a multipoint linkage study of the fragile X syndrome. 35 Australian pedigrees were genotyped for the three new RFLPs and other nearby polymorphisms. Genotypings were obtained from a further 77 pedigrees as part of an international collaborative project. On multipoint linkage analysis of these data, FRAXA was located 2 cM proximal to VK21. These results define a new diagnostic strategy for DNA studies of fragile X families. A combination of five probes and three restriction endonucleases will identify an RFLP within 4 cM of FRAXA in 94% of women.

The genetic maps derived from the normal families and from the fragile X families were compared. Contrary to previous reports, there was no difference in these genetic maps between or within the populations.