gene is linked to C-ets 1 on the long arm of chromosome 11 in these families. More families are being studied to confirm the linkage of NF to C-ets 1.

References

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Genetic linkage studies with neurofibromatosis: the question of heterogeneity

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SUMMARY: Three new families are reported for standard gene linkage markers and classical peripheral neurofibromatosis (Von Recklinghausen disease). Additional data are summarised for the exclusion map. One family gives slight evidence of close linkage with the Gc locus on chromosome 4, raising again the question of possible genetic heterogeneity in NF.

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The families in this report have not been included in our earlier studies.1,2 Diagnoses were made by the same criteria as reported earlier, and all of the families were segregating classical peripheral neurofibromatosis. There were no cases of reduced penetrance in the three families, although there were subjects still in the age at risk to develop NF.

Materials and methods

The three families were ascertained through a nationwide search for families with multiple affected subjects conducted by the Clinical Epidemiology Branch, National Cancer Institute. Family (UCLA ID number) NF024 had marker studies performed on nine subjects, NF026 on six subjects, and NF028 on 27 subjects. In addition, NF028 had two brothers married to two sisters. Laboratory studies for the gene markers were carried out using standard techniques.1,2 Lod scores were calculated using the computer programme LIPED with a straight line age of onset correction going from 0 penetrance at 10 years to 98 at 30 years.3 The affected allele frequency was set at 0.001.

Results

The additional exclusion data available from the analyses of these three families, as well as a summary of the positive scores, are presented in the table. Analyses are completed on family NF028 only for the Gc locus on chromosome 4, due to time constraints. For all analyses the lod scores are shown only for a recombination frequency of 0-01 in males and females, since these values represent the maximum lod scores for those markers with positive scores and also give the strongest evidence against linkage for the other markers. The families were also informative for the P blood group marker although the combined score was only -0.21 for the two families.

Discussion

The additional data presented here will assist in extending the exclusion map for the NF locus. These three families continue the pattern described by the reports,1,2 most of our families show strikingly negative scores with Gc. Families NF024 and NF026 repeated that pattern in this report. However, when we have families with positive lod scores, the maximum occurs at the close recombination value of 0.01. This differs from the pattern in that random positive scores expected if the two loci are not linked but tend to cosegregate to varying degrees in families by chance alone. A definitive test for a proportion of families to be linked to Gc has not yet been accomplished, as the total number of families typed for the marker and available to test remains small.

The exclusion map is growing rapidly for NF and the data will soon be available to explore the remaining regions not yet excluded. A possible outcome from this cooperative effort is that the entire genome will be excluded, bringing about confirmation of heterogeneity in an indirect manner not dreamed possible a few years ago. However, it may be possible to show heterogeneity more directly and sooner by pooling families from all published studies and performing the appropriate heterogeneity test.

References

DNA linkage analysis in Von Recklinghausen neurofibromatosis


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SUMMARY We have used DNA linkage analysis in 11 families with Von Recklinghausen neurofibromatosis (VRNF) in order to search for the chromosomal localisation of the defective gene causing this serious neurological disorder. Three groups of polymorphic DNA markers were used: (1) markers for chromosome 22, because of possible allelic genetic heterogeneity between VRNF and bilateral acoustic neurofibromatosis; (2) markers near the centromere of chromosome 4, since there was preliminary evidence for linkage between the VRNF gene and Gc; and (3) oncogenes and growth factors as possible candidate genes for VRNF. Our data exclude close linkage between any of these markers and the gene for VRNF.

DNA linkage analysis was performed in 11 families with Von Recklinghausen neurofibromatosis (VRNF) according to diagnostic criteria which are in agreement with those recently established by NIH. Seven of these families have been described previously, and the other four families are among those described by Upadhyaya et al in this issue. Blood samples from 161 subjects were collected, comprising 80 samples from affected persons and 79 potentially informative meioses. Lymphocytes were isolated from these blood samples and transformed into permanent lymphoblastoid cell lines by Epstein-Barr virus. DNA was isolated and digested with appropriate restriction enzymes. The resulting DNA fragments were separated according to their molecular weights by gel electrophoresis, transferred to nylon membrane, and hybridised to radiolabelled DNA probes which were known to reveal restriction fragment length polymorphisms (RFLPs) in human genomic DNA. DNA linkage analysis was performed by use of the linkage programme LIPED, assuming a penetrance of the defect of 95%.

As shown in the table, three groups of polymorphic DNA markers were used for linkage analysis. (1) Markers for chromosome 22, since acoustic neuromas and meningiomas from patients with bilateral acoustic neurofibromatosis show highly specific deletions on this autosome, suggesting that the gene causing bilateral acoustic neurofibromatosis is located on chromosome 22. Furthermore, Krone and Hogemann reported in a recent cytogenetic study that cultured peripheral neurofibromas from patients with VRNF were associated with various chromosomal aberrations, the most consistent of which was monosomy for chromosome 22. (2) Markers near the centromere of chromosome 4 (ALB, D4S1, and D4S35), since there was preliminary evidence for linkage between Gc on chromosome 4 and VRNF. (3) Oncogenes, growth factors, and their receptors as possible candidate genes for VRNF, a disease which is associated with tumour formation. However, the linkage data presented in the table exclude close linkage between any of these DNA markers and the gene for VRNF.