Linkage of the tuberous sclerosis locus to a DNA polymorphism detected by *v-abl*

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SUMMARY Linkage analysis was undertaken in six families with tuberous sclerosis (TS) using a restriction fragment length polymorphism detected by *v-abl*. No recombinants were observed in 13 informative meioses (four phase known) giving a maximum lod score of 3.18 at zero recombination (confidence limits 0 to 0.15). This provides further evidence for the assignment of TS to 9q34 and should facilitate cloning of the structural gene, genetic counselling, and first trimester prenatal diagnosis.

Tuberous sclerosis (TS) is an autosomal dominant disorder with a variable phenotype which commonly includes hypopigmented skin macules, adenoma sebaceum, ungual fibromas, seizures, and mental retardation.¹ The initial suggestion of linkage of TS to the ABO blood group² has recently been confirmed in a collaborative UK study.³ This maps the locus for TS to 9q34. We provide here further evidence for this assignment.

Methods

Six families with TS suitable for linkage analysis

were ascertained from the genetic records of the West of Scotland Regional Genetics Centre. All affected subjects conformed to the diagnostic criteria of Gomez¹ and unaffected subjects at risk were rigorously investigated as detailed elsewhere.³

The methods for DNA extraction and restriction fragment identification have been described previously.⁴ DNA from each family member was digested with *TaqI* and after electrophoresis and Southern blotting was probed with pSA-19 which is a 1.9 kb fragment of *v-abl*.⁵ Lod scores were calculated using the computer programme LIPED⁶ assuming a penetrance of 98%⁷ and the confidence interval was determined by taking values of the recombination fraction corresponding to a lod score one unit less than the maximum.⁸

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FIG 1 Abbreviated pedigrees of informative TS families. Numbers above symbols correspond to lanes in fig 2.

FIG 2 *TaqI* digested DNA probed with *v-abl* (pSA-19). Polymorphic bands are arrowed and lane numbers correspond to the subjects shown in fig 1.

Results

Three of the six families were informative for this RFLP and their pedigrees and DNA autoradiographs are shown in figs 1 and 2. Subjects 6, 13, 14, 27, and 29 had normal clinical and ophthalmological examinations (including examination of the skin under ultraviolet light) but were excluded from the analysis because they had not had cranial CT scans, renal ultrasound, and *x* rays of the hands and feet, as demanded by the rigorous protocol adopted for an earlier study.³ With these exclusions no recombinations were observed in 13 informative meioses giving a peak lod score of 3.18 at zero recombination (table). If these subjects had been included as unaffected there would have been no recombinations in 18 informative meioses giving a maximum lod score of 4.64 at zero recombination.

Discussion

The human oncogene *c-abl* is the cellular homo-

logue of the transforming sequence *v-abl* which was first discovered in the Abelson strain of murine leukaemia virus (A-MuLV). The coding sequences for *c-abl* extend over a 225 to 300 kb region and the *v-abl* homologous regions (VAHR) are located at its 3' end.⁹ Human *c-abl* has been localised to 9q34 by in situ hybridisation and somatic cell hybrid panels and is transferred to chromosome 22 in the reciprocal translocation which creates the Philadelphia chromosome.¹⁰⁻¹⁴ The adenylate kinase 1 (AK1) linkage group has also been regionally localised to 9q34 by dosage studies using children with various imbalances of chromosome 9,¹⁵ but unlike *c-abl* is not transferred to chromosome 22 in the Philadelphia chromosome¹⁶ which would suggest that *c-abl* is distal to AK1.

Using *c-abl* (ablK-2) Barker and White¹⁷ have described diallelic polymorphisms with *TaqI*, *PstI*, *AccI*, and *StuI* (each with an infrequent allele frequency of 0.11). The present study using the *TaqI* polymorphism detected by *v-abl* has shown linkage to TS and provides further evidence for the assignment of TS to 9q34. This marker will be more useful for early prenatal diagnosis than the ABO or AK1 loci and will aid attempts to clone the structural gene for TS.

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TABLE Lod scores at different values of the recombination fraction (θ) for tuberous sclerosis versus *v-abl*.

Recombination fraction (θ)							$\hat{Z}$	$\hat{\theta}$	Confidence limit for θ
0	0.05	0.1	0.2	0.3	0.4				
3.18	2.85	2.52	1.83	1.13	0.47	3.18	0	0-0.15	

$\hat{Z}$ =maximum lod score. $\hat{\theta}$ =corresponding value of recombination fraction.

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Note added in proof

Since submission, first trimester prenatal exclusion has been performed using this RFLP for subject 12 in GLA 4136 and his wife.