Genetic aspects of tuberous sclerosis in the west of Scotland

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SUMMARY Complete ascertainment of tuberous sclerosis was attempted in the west of Scotland (population 2 763 000). A total of 101 patients was identified, giving an overall minimum prevalence of 1 in 27 000, but for children under 10 years of age the minimum prevalence was 1 in 12 000. Both parents of 84 of the ascertained cases were assessed for signs of tuberous sclerosis. In 51 pairs of parents no evidence of the condition was seen, indicating that up to 60% of the cases were new mutations. The mutation rate was estimated at 2.5×10^{-5} mutations per gene per generation. Analysis of parental ages for the new mutations did not show a significant age effect. Thirty-five patients occurred in 13 families containing other affected subjects. The pattern of inheritance was consistent with an autosomal dominant trait in these families. In one sibship non-penetrance or gonadal mosaicism resulted in affected sibs with normal parents. Of two further sibships where non-penetrance was suspected, one was shown to represent a single new mutation in monozygotic twins and the other to involve non-paternity.

Tuberous sclerosis (TS) is an accepted autosomal dominant trait with characteristic dermatological and neurological features (table 1). Mental handicap occurs in more than half of recognised affected subjects.¹ The variable expression seen in TS can often complicate initial diagnosis; this, however, is facilitated by adherence to diagnostic criteria as described by Gomez.² In recent years the recognition of more diagnostic signs together with more complete evaluation of family members has led to increasing estimates of the prevalence of TS.¹ The occasional inability to detect any diagnostic features in obligate carriers of the TS gene has led to reports of non-penetrance or gonadal mosaicism³⁻⁵ with implications for genetic counselling of the families of apparently sporadic cases. The present study aimed to determine the prevalence and to investigate the main genetic aspects of tuberous sclerosis in the west of Scotland.

Patients, methods, and results

ASCERTAINMENT

TS patients were ascertained from the records of the

West of Scotland Regional Genetic Service, the Fraser of Allander Assessment Unit (paediatric neurology), and the Tuberous Sclerosis Association.

Consultants in paediatrics, neurology, dermatology, and mental handicap in the region (approximately 100) were approached by questionnaire in order to maximise ascertainment and all responded. Of the 101 cases identified, 97 were seen by one or more of the authors; the other seven had been diagnosed by other hospital specialists. The diagnostic criteria of Gomez² (table 1) were fulfilled by all

TABLE 1 Diagnostic criteria for tuberous sclerosis (after Gomez²).

Primary criteria (only one required for diagnosis)	Secondary criteria (two required for diagnosis)
Adenoma sebaceum	Infantile spasms
Ungual fibromas	Hypopigmented macules
Cortical tuber	Shagreen patch
Subependymal hamartomas	Single retinal hamartoma
Multiple retinal hamartomas	Bilateral renal cysts or angiomyolipomata
Fibrous forehead plaque	Cardiac rhabdomyoma(ta)
	First degree relative with TS

The pulmonary and gastrointestinal systems are occasionally involved.

patients in the study. Where possible both parents (and usually sibs) were assessed for signs of TS.

PREVALENCE

A total of 101 cases, 52 male and 49 female, was identified living in the west of Scotland, population 2 763 000,⁶ on the study date of 1 June 1986. The overall prevalence of ascertained cases was 1 in 27 000 and in the population under 10 years 1 in 12 000. The birth frequency is predicted to be higher than the latter figure as mildly affected subjects will often not be diagnosed during childhood.

INHERITANCE

Both parents of 84 of the 101 patients in the study were clinically assessed for signs of TS (this included fundoscopy and Wood's light examination of the skin). In the remainder, assessment was not possible owing to the death of one or both parents or separation with one being untraceable. In two families the affected children had been adopted. In 51 of the 84 cases there was no evidence of TS in either parent. Thus, the proportion of sporadic cases in this group was 60%. If all those cases in which examination of both parents was not possible represented new mutations, the overall proportion of new mutations could be as high as 70%.

Thirty-five patients were members of families with more than one affected subject. In one family four generations were affected, in four families three generations, and in seven families two generations. In these families TS was inherited as an autosomal dominant trait and no skipped generations were seen. Great variation in expression was evident within as well as between families. Subgrouping of clinical features among these families, which might suggest heterogeneity, was not noted.

A set of affected twins with normal parents (Glasgow pedigree 4521) have previously been reported as dizygotic on the basis of serum alanine

aminotransferase phenotypes.⁷ DNA fingerprinting, however, showed them to be monozygotic and to represent a single new mutation. In a second pedigree, previously considered to illustrate non-penetrance (Glasgow pedigree 3421⁵), the α globin 3' hypervariable probe was used to show non-paternity. Inheritance from the biological father could not be confirmed as he was not available for clinical assessment. The parents of a further pair of affected sibs (Glasgow pedigree 4151) were assessed clinically (including Wood's light and fundoscopy), by brain CT scan and renal ultrasound, all with normal results. Paternity was confirmed using the α globin 3' hypervariable probe. This was the only pedigree in which an obligate gene carrier did not fulfil the diagnostic criteria of Gomez.²

PARENTAL AGE EFFECT

For sporadic cases the mean paternal age of 30.02 years (SD 6.6) and mean maternal age 27.2 years (SD 5.7) did not differ significantly from those in the Scottish population (paternal 30.8 years, SD 7.07; maternal 27.2 years, SD 6.23, calculated from the Report of the Registrar General for Scotland 1969) ($p>0.1$, Student's *t* test).

MUTATION RATE

Assuming complete penetrance, a birth frequency of 1 in 12 000, and the proportion of new mutations to be at least 60%, a minimum estimate of the mutation rate is 2.5×10^{-5} mutations per gene per generation.

Discussion

The overall prevalence of tuberous sclerosis found in this study was 1 in 27 000, slightly higher than previous estimates based on large numbers of cases (table 2). Early population surveys greatly underestimated the prevalence of TS.⁸⁻¹¹ Other authors

TABLE 2 Tuberous sclerosis: previous genetic studies.

Authors	Reference	Cases	Estimated prevalence	Proportion of sporadic cases	Mutation rate ($\times 10^{-5}$)
Gunther and Penrose	12	20	1 in 30 000	50%	16
Borberg	8	37	—	57-79%	—
Stevenson and Fisher	9	9	1 in 150 000	7 of 9	—
Nevin and Pearce	11	18	1 in 100 000	66%	10.5
Zaremba	13	40	1 in 23 000	58%	11
Bundey and Evans	17	71	—	86%	—
Singer	10	26	1 in 170 000	65%	6
Donegani <i>et al</i>	14	6	1 in 10 000	—	—
Fleury <i>et al</i>	16	48	—	56%	—
Hunt and Lindenbaum	1	68	1 in 34 000	50-75%	25
Wiederholt <i>et al</i>	15	8	1 in 9400	6 of 8	—
Present study		104	1 in 27 000	60%	25

studied subpopulations of institutionalised patients^{12 13} or based their estimates on very small numbers of cases.^{14 15} The recognition of further diagnostic signs and formulation of diagnostic criteria² has enabled better ascertainment in recent studies.^{1 16} Estimates of the prevalence of TS will inevitably be underestimates because of the occurrence of minimally affected cases who may not come to medical attention. We often ascertained mildly affected subjects through more severely affected descendants. As in previous studies a much higher prevalence was found among children (1 in 12 000 under 10 years) than among adults. This reflects in part the increased mortality associated with the condition and possibly increasing diagnostic awareness among paediatricians.

Because most large studies have not attempted thorough assessment of the parents of apparently sporadic cases, the proportion of new mutations is likely to have generally been overestimated. Although we attempted to examine the parents of index cases wherever this was possible, they were only rigorously investigated (brain CT scan, renal USS, skeletal survey) where further children were being considered, doubtful signs of TS were found, or if they were taking part in linkage studies. The proportion of sporadic cases among those whose parents were clinically evaluated is compared with previous reports in table 2. The lack of a significant parental age effect for new mutations is in keeping with the findings of previous studies which have addressed this question.^{8 11 17} The mutation rate for the TS gene has most recently been estimated at 2.5×10^{-5} per gene per generation.¹ We derived the same figure from our data, which gave a higher prevalence but a lower proportion of fresh mutations. This mutation rate would suggest 18 new cases of TS per year to normal parents in the UK.

Among those families in which more than one person was affected, only one subject was thought to be a gene carrier not fulfilling Gomez's criteria.² Affected sibs with normal parents, apparent gene carriers, are described elsewhere,^{3 5} as is a pedigree in which affected cousins have normal parents.⁴ In other reported families presumed gene carriers have been incompletely investigated.^{18 19} Non-penetrance, gonadal mosaicism, genetic heterogeneity (recessive inheritance), and non-paternity might account for the observation of these unusual pedigrees. This has practical relevance and when counselling the normal parents of apparently sporadic cases we currently advise a recurrence risk of 1%. Although ultrasound detection of fetal cardiac rhabdomyomata has enabled prenatal diagnosis of TS,^{20 21} its reliability is unknown.

Linkage between TS and the ABO blood group

has been reported,²² suggesting that the TS locus (TSC) maps to the distal long arm of chromosome 9. A further study found linkage between a polymorphism at the *c-abl* locus, which maps to chromosome 9 band q34, and TS.²³ Subsequently, early prenatal exclusion was reported in an extensive family showing linkage in that study.²⁴ Recently, recombinations between *ABO* and *TSC* have been noted^{25 26} and the question of genetic heterogeneity has been raised. In the families ascertained in the present study there was no clinical evidence of heterogeneity, but further linkage studies are clearly required before prenatal DNA diagnosis can be more widely offered.

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