Phenotype-genotype correlations in X linked retinitis pigmentosa

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Abstract

Retinitis pigmentosa (RP) represents a group of clinically heterogeneous retinal degenerations in which all modes of inheritance have been described. We have previously found two different clinical profiles in X linked RP as a function of age and mode of onset. The first clinical form has very early onset with severe myopia. The second form starts later with night blindness with mild myopia or none. At least two genes have been identified in X linked forms, namely RP2 (linked to *DXS7*, *DXS255*, and *DXS14*) and RP3 (linked to *DXS84* and *OTC*) on the short arm of the X chromosome.

In order to contribute to phenotype-genotype correlations in X linked RP, we tested the hypothesis that the two clinical profiles could be accounted for by the two different gene loci. The present study provides evidence for linkage of the clinical form with early myopia as the onset symptom with the RP2 gene (pairwise linkage to *DXS255*: $\hat{Z}=3{\cdot}13$ at $\hat{\theta}=0$), while the clinical form with later night blindness as the onset symptom is linked to the RP3 gene (pairwise linkage to *OTC*: $\hat{Z}=4{\cdot}16$ at $\hat{\theta}=0$).

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Retinitis pigmentosa (RP) is a progressive hereditary disorder that primarily alters photoreceptor and pigment epithelial function.[1,2] It represents a group of clinically distinct retinal degenerations and all modes of inheritance have been described.[3–7]

We have recently identified two different clinical profiles in X linked RP (XLRP) as a function of age and mode of onset of the disease. The first has very early onset with myopia (mean age of onset (1 SD) = 3·5 (0·5) years) and the second starts later with night blindness (mean age of onset (1 SD) = 10·6 (4·1) years[8]). At least two loci have been identified in XLRP, namely *RP2* (Xp11.21-p11.4.1) and *RP3* (Xp21.1),[9–16] but all previous reports have failed to correlate either gene locus with any clinical presentation in this group.[17–19]

Considering the evidence for two clinical subtypes on one hand and two different loci on the other, we decided to test the hypothesis that the clinical heterogeneity of XLRP could be accounted for by the two different loci on the short arm of the X chromosome. The present study provides evidence for the correlation of each clinical subtype with each particular gene locus and shows that the clinical form with early myopia is accounted for by the RP2 gene, while the clinical form with later night blindness is accounted for by the RP3 gene.

Patients and methods

PATIENTS

Nine XLRP families were identified in five different medical genetic centres in France. X linked inheritance was shown by maternal transmission over at least two generations and absence of male to male transmission. Our diagnostic criteria for RP were those laid down by the 1982 International Symposium of Ophthalmology[1]: (1) bilateral involvement, (2) concentric depression of the visual field, (3) severe scotopic involvement on electroretinogram (ERG), resulting from alteration of rods, or no ERG response, and (4) progressive loss of photoreceptor function.

These nine families were split into two groups as a function of age and mode of onset of the disease as we have previously reported.[8] In the first group (XLRP-A, families 1 to 5, figs 1 to 5), affected boys presented with typical RP. They first mentioned night blindness at around 10 years of age, then they developed a gradual constriction of the visual field with extinction of their ERG. Their visual acuity was preserved until 15 years of age and rapidly declined thereafter (table 1). The five pedigrees included 11 available affected males, six available healthy males, 10 obligate carriers, and eight potential carriers. Among the obligate carriers, only eight women accepted ophthalmological examination. Two carriers had no clinical symptoms, while six had clinical symptoms, including tapetal-like reflex (metallic sheen, three cases), sectorial pigmentary deposits (three cases), or both (two cases). Among the eight potential carriers, two had sectorial pigmentary deposits and one had both pigmentary migration and metallic sheen.

In the second group (XLRP-B, families 6 to 9, figs 6 to 9), affected boys developed an early onset, progressive, high myopia (mean age 0 to 3 years, table 1). They had had reduced visual acuity since nursery school while night blindness and visual field constriction was first noted at 10 to 14 years only. The ERG was extinguished very early. After 14 years of age, the fundus displayed non-specific abnormalities including pigmentary deposits, narrowed vessels, and optic nerve pallor (table 1). The four pedigrees included eight available affected males, three available healthy males, eight

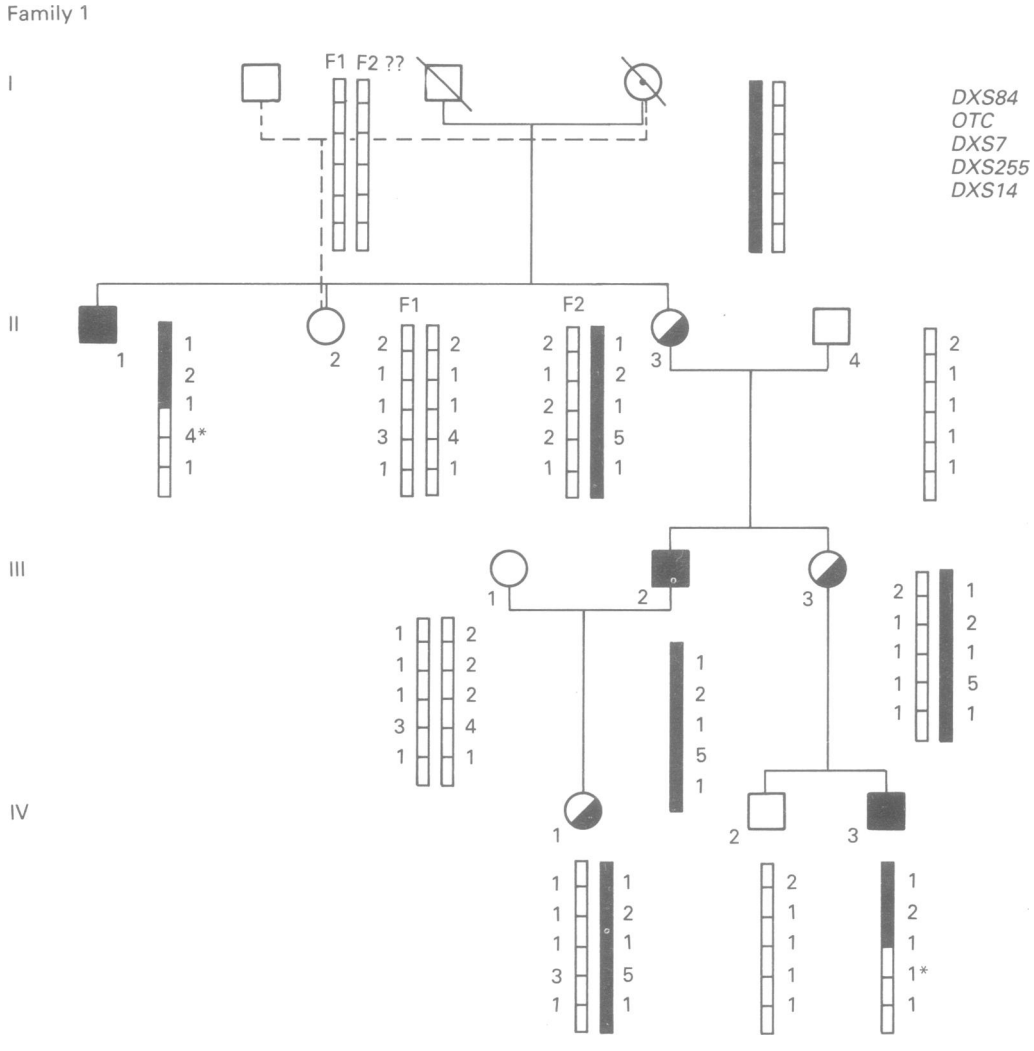


Figure 1 Segregation of RFLP alleles with XLRP in family 1. The summary of the probes and alleles is given above in the same order as the data presented along the sides of the chromosome stick figures. Asterisks mark the loci that are recombined with XLRP. II·1 and IV·3 are recombinant for DXS255.

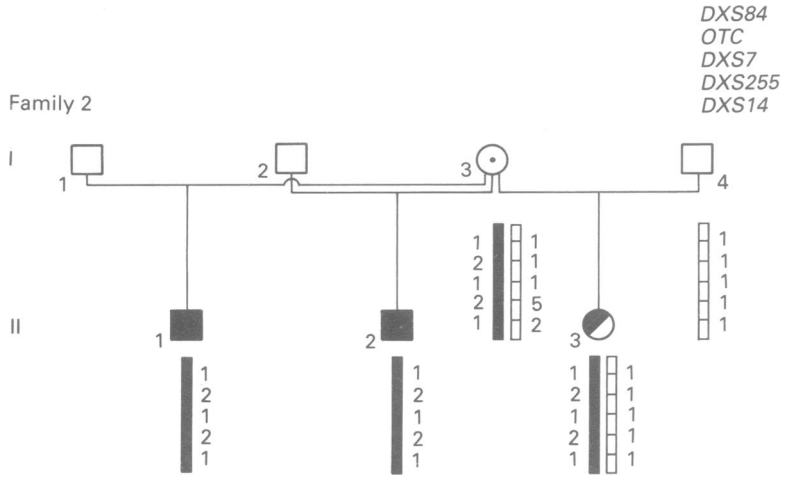


Figure 2 Segregation of RFLP alleles with XLRP in family 2.

obligate carriers, and eight potential carriers. Of the eight obligate carriers, only four accepted ophthalmological examination. One had a normal fundus but a delayed scotopic response, two had a typical tapetal reflex, and one had sectorial and peripheral pigment deposits on her fundus and ERG showed scotopic disturbance. Of the eight potential carriers, one had a normal fundus but a delayed scotopic response and three displayed sectorial pigmentary deposits in their retina without tapetal-like reflex. These three women did not accept electrodiagnostic examination (table 1).

Methods

For restriction fragment length polymorphism (RFLP) analysis, total DNA was extracted from circulating leucocytes by cell lysis, proteinase K digestion, phenol/chloroform extraction, ethanol precipitation, and Tris-EDTA resuspension. DNA (5 μg) was then cleaved with restriction enzymes *Pst*I, *Msp*I, or *Taq*I under appropriate buffer and temperature conditions according to the manufacturer's recommendations. The fragments were separated by horizontal gel electrophoresis in Tris-acetate EDTA buffer and transferred onto a nylon membrane (Zetabind, Flo Cuno)

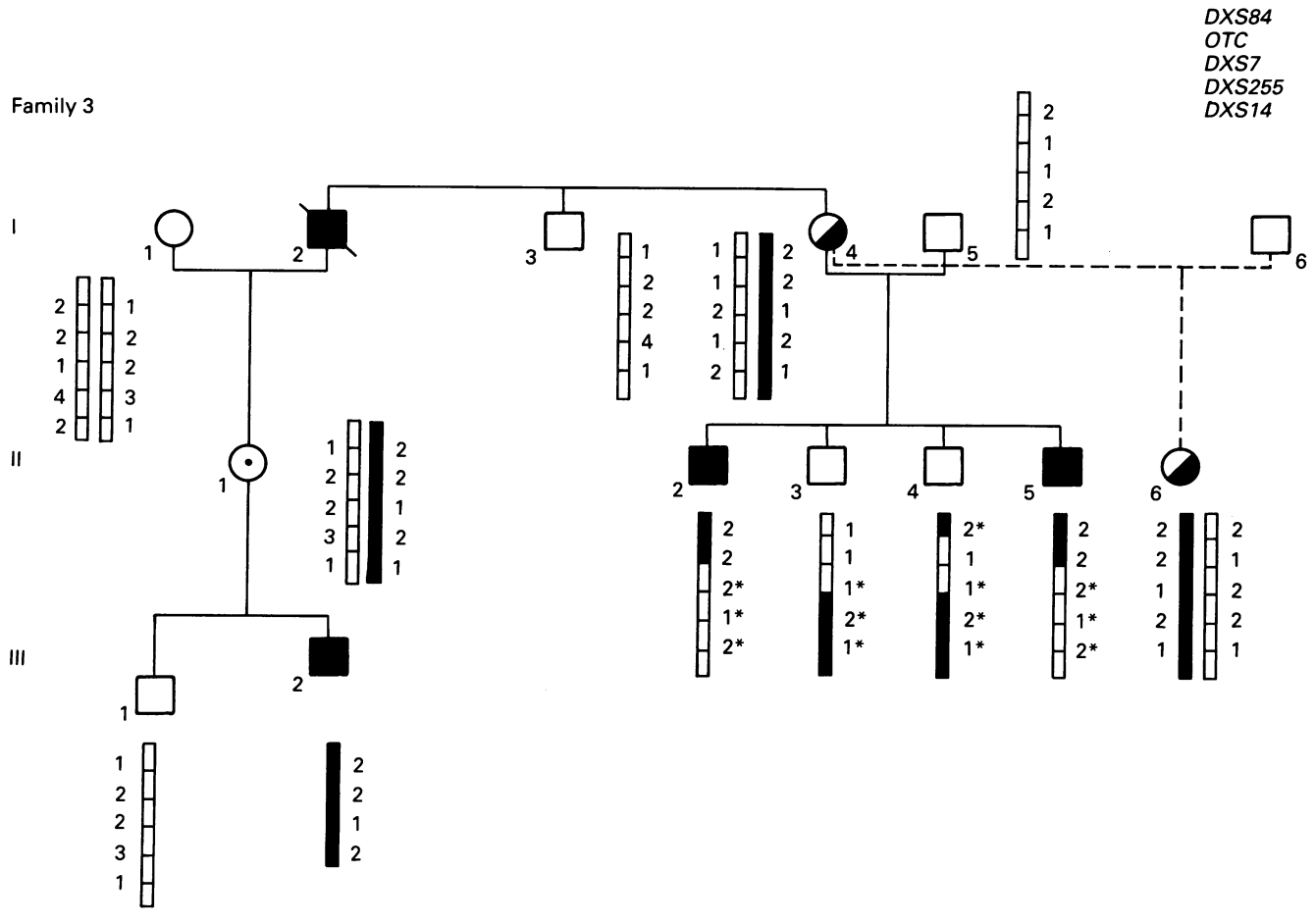


Figure 3 Segregation of RFLP alleles with XLRP in family 3. II·2, II·3, and II·5 were recombinants for DXS7, DXS255, and DXS14. II·4 was recombinant for DXS84, DXS7, DXS255, and DXS14. III·2 was an affected male fetus, analysed after chorionic villi sampling. The pregnancy was terminated.

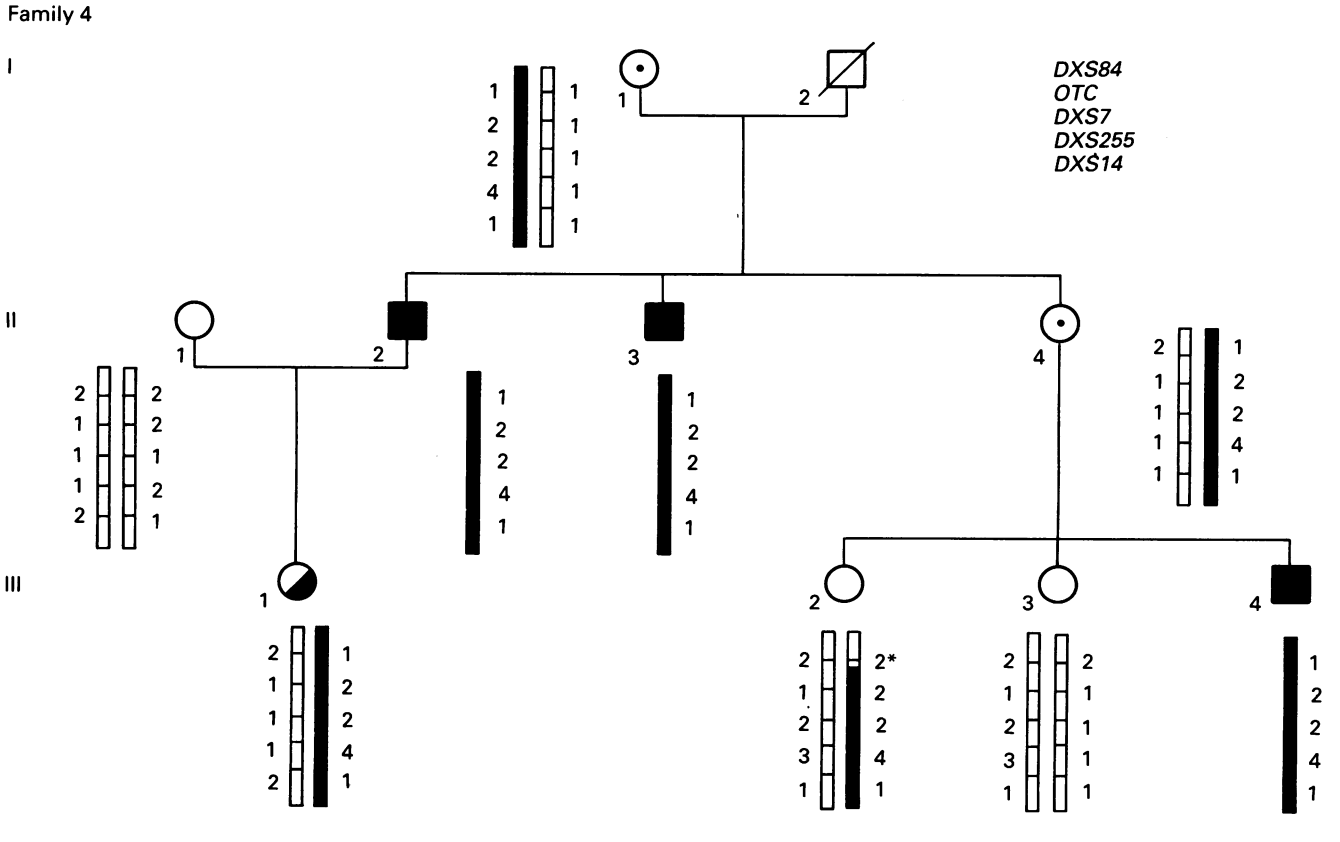


Figure 4 Segregation of RFLP alleles with XRLP in family 4. III·2 was recombinant for DXS84.

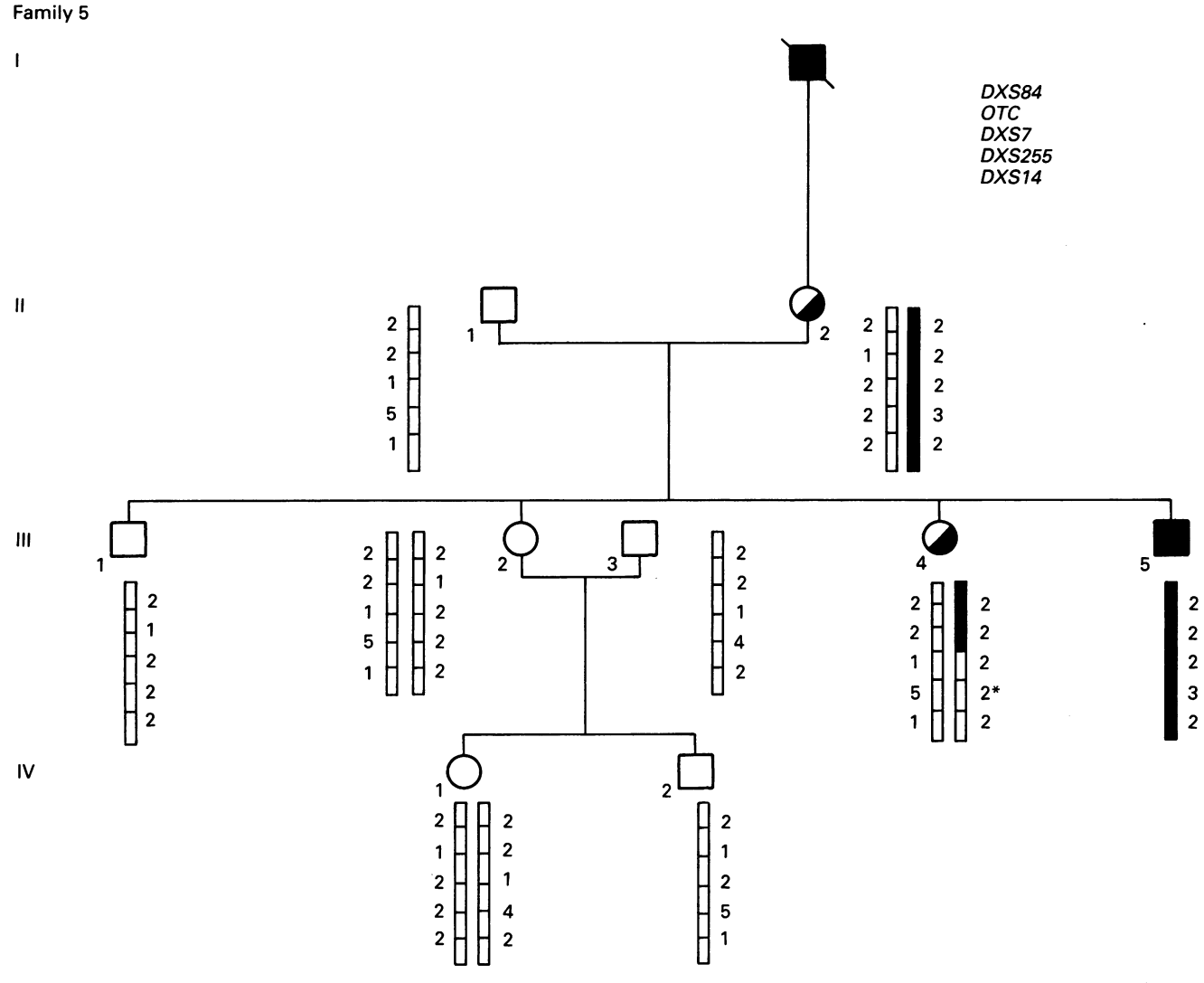


Figure 5 Segregation of RFLP alleles with XLRP in family 5. III·4 was recombinant for DXS255.

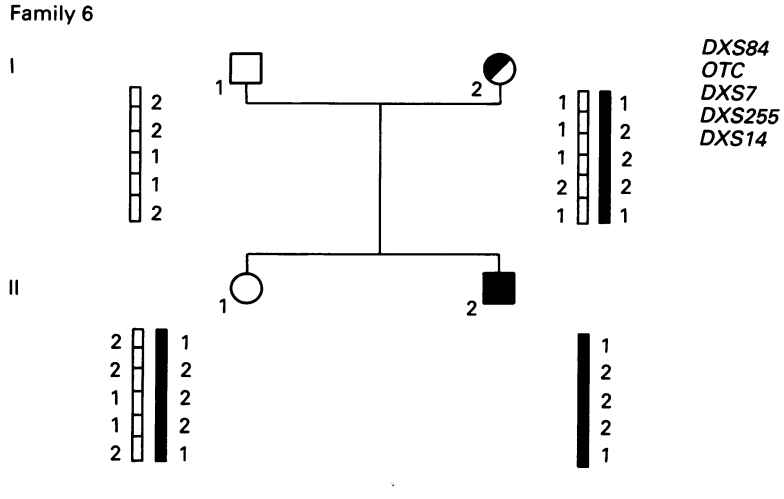


Figure 6 Segregation of RFLP alleles with XLRP in family 6.

using Southern's technique. Probes were radiolabelled by nick translation to a high specific activity with ^{32}P-dCTP.[20] After hybridisation, the filters were washed and exposed to Kodak X/OMAT films with intensifying screens. The polymorphic loci studied were Xp2.1—*DXS84*$^{6\,cM}$*OTC*$^{11\,cM}$*DXS7*$^{10\,cM}$ *DXS255*$^{9\,cM}$*DXS14*—Xp1.1 (table 2).

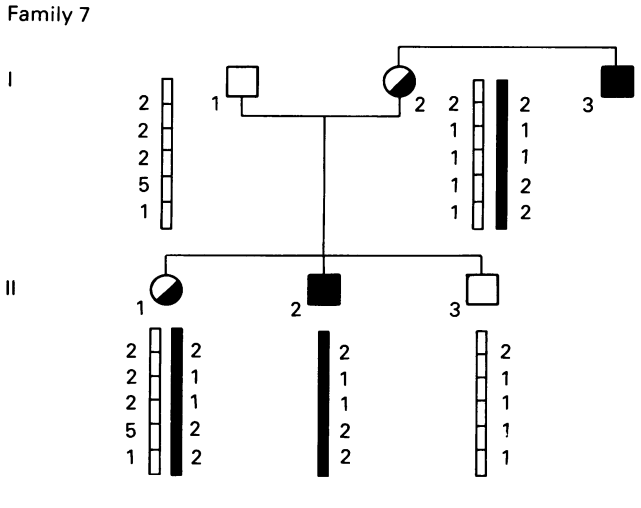
Linkage analysis was performed using the LINKAGE program version 4·8[21] on an IBM PC AT. Two point linkage analysis was performed between the RP locus and each marker using the MLINK option. Multipoint linkage analysis was performed using the LINKMAP program.[22] Genetic distances between markers were obtained from previous studies.[18 23–25] Multipoint lod scores were obtained by moving the disease locus between the markers and the genetic map was constructed by calculating the lod score for a given map distance.

The hypothesis of linkage homogeneity H1 specifies $\theta_1 = \theta_2 < 1/2$. The alternative hypothesis of linkage heterogeneity H2 is given by $\theta_1 \neq \theta_2$, where the recombination fraction is potentially different in each family class. To test H1 against H2, we used the Morton likelihood ratio (LR) test, where Z_1 (θ_1) denotes the total lod score of the first class and Z_2 (θ_2) the total lod score of the second class for a value of

Table 1 Ocular findings in families with RPLX.

Family	Patient	Status	Age	Mode of onset	Age of onset	Age of NB	Refractive error (D)	Fundus abnormalities	Visual field abnormalities	Present ERG	Age of decreased VA	Present VA
1	II·1	Affected	55	NB	10	10	?	?	?	?	17	Blind
	III·2	Affected	33	NB	5	5	−6	Pigmentary deposits all over the retina, narrowed vessels, macular degeneration	Major constriction	Extinguished	18	1/20
	II·3	Obligate carrier	52	—	—	—	Emmetropia	Tapetal reflex	Normal	Scotopic alteration	Normal	Normal
	III·3	Obligate carrier	28	—	—	—	Emmetropia	Tapetal reflex	Normal	Normal	Normal	Normal
	IV·1	Obligate carrier	2	—	—	—	Emmetropia	Irregularity of pigmentation	Not done	Normal	Normal	Normal
	IV·3	Affected		Not examined								
2	II·1	Affected	35	NB	9	9	Emmetropia	Pigmentary deposits, narrowed vessels	Major constriction	Extinguished	About 20	Blind
	II·2	Affected	31	NB	8	8	Emmetropia	Pigmentary deposits, narrowed vessels	Major constriction	Extinguished	About 20	2/20
	I·3	Obligate carrier	59	—	—	—	Emmetropia	Normal	Normal	Not done	Normal	Normal
	II·3	Potential carrier	27	—	—	—	Emmetropia	Sectorial pigmentary deposits in right eye	Localised scotoma in RE corresponding to pigments	Not done	Normal	Normal
3	II·2	Affected	25	NB	8	8	Astigmatism	Pigmentary deposits all over the retina, narrowed vessels	Big annular scotoma	Extinguished	About 18	2/20
	II·5	Affected	18	NB	5	5	Emmetropia	Pigmentary deposits all over the retina, narrowed vessels	Constriction	Extinguished	18	4/10
	II·1	Obligate carrier	30	—	—	—	Emmetropia	Normal	Normal	Not done	Normal	Normal
	I·4	Obligate carrier	56	—	—	—	Astigmatism	Sectorial deposits in the retina	Mild constriction	Not done	After 40	10/20
	II·6	Potential carrier	17	—	—	—	Astigmatism	Sectorial deposits in RE	Normal	Not done	—	Normal
4	II·2	Affected	60	NB	6–8	6–8	Emmetropia	Pigmentary deposits, RP at a terminal stage	Major constriction	Extinguished	20	Blind
	II·3	Affected	56	NB	6–8	6–8	Emmetropia	Not examined		?	20	Blind
	III·4	Affected	19	NB	#6	#6	Emmetropia	Pigmentary deposits, narrowed vessels	Constriction	Extinguished	18	4/20
	II·4	Obligate carrier	48	—	—	—	—	Not examined		Not done	—	?
	III·1	Obligate carrier	34	Mild NB	Teenage	Teenage	−16LE −2RE	Tapetal reflex, sectorial pigment	Mild constriction	Scotopic alteration	—	LE 6/20 RE 20/20
	III·2	Potential carrier	24	Not examined								
5	III·5	Affected	27	NB	5	5	−3	Pigmentary deposits, retinal atrophy	Constriction	Extinguished	17	2/20
	II·2	Obligate carrier	55	—	—	—	Astigmatism	Tapetal reflex, localised pigmentary deposits in LE	Scotoma in LE corresponding to pigment	Normal	?	10/20
	III·4	Potential carrier	30	—	—	—	Emmetropia	Tapetal reflex and pigment on periphery of retina in RE	Scotoma in RE corresponding to pigment	Not done	Normal	Normal
6	II·2	Affected	19	High myopia	4	10	−10 −15	Pigmentary deposits all over the retina, narrowed vessels and optic atrophy	Major constriction	Extinguished	17	2/20
	I·2	Potential carrier	53	—	—	—	Emmetropia	Sectorial deposits in RE	Not done		—	18/20
7	II·2	Affected	22	High myopia	2	10	−18	Pigmentary deposits, narrowed vessels, degenerative macula	Major constriction	Extinguished	15	2/20
	I·2	Obligate carrier	43	—	—	—	Emmetropia	Normal	Normal	Delayed scotopic response	—	Normal
	II·1	Potential carrier	24	—	—	—	−1RE −2LE	Normal	Normal	Delayed scotopic response	—	Normal
8	II·5	Affected	53	High myopia	2	7	−10	RP at a terminal stage	Major constriction	Not done	20	Blind
	III·2	Affected	41	High myopia	Early infancy	?	−10	?	?	?	20	Blind
	IV·2	Affected	7	High myopia	3	?	−8·50	Reduced vessels, irregularity of retina	Annular scotoma	Extinguished	6	2/20
	IV·3	Affected	4	High myopia	1	?	−10	?	Annular scotoma	Extinguished	?	?
	III·7	Obligate carrier	30	—	—	—	Emmetropia	Tapetal reflex	Normal	Not done	—	Normal
	IV·1	Potential carrier		Not examined								
9	II·4	Affected	58	High myopia	6	13	−10	Pigmentary deposits, degenerative macula	Major constriction	Not done	16	Blind
	III·2	Affected	30	High myopia	3	14	−6	Pigments all over the retina	Major constriction	Extinguished	16	2/20
	II·1	Potential carrier	62	—	—	—	Astigmatism	Sectorial pigmentary deposits	Normal	Not done	Normal	16/20
	II·2	Obligate carrier	60	—	—	—	Hyperopia	Tapetal reflex	Normal	Not done	Normal	Normal
	III·1	Potential carrier	33	—	—	—	Hyperopia	Sectorial deposits in one eye	Normal	Not done	Normal	Normal
	III·4	Obligate carrier	23	—	—	—	Emmetropia	Sectorial deposits in RE, peripheral pigments in LE	Normal	Scotopic alteration	Normal	Normal

NB = night blindness. ERG = electroretinogram. VA = visual acuity. D = dioptres. LE = left eye. RE = right eye.



θ at which Z is largest.[26] The log of the LR statistic is then given by $\chi^2 = 2(\ln 10)\, Z_1(\theta_1) - Z(\theta) + Z_2(\theta_2) - Z(\theta)$. The test is considered significant and homogeneity is rejected when χ^2 is larger than the appropriate critical χ^2 value with two degrees of freedom.

As our clinical classification is based on subjective criteria, we also performed the test HOMOG, version 2·4,[22] on the total data derived from all nine families, ignoring their clinical presentation.

Figure 7 Segregation of RFLP alleles with XRLP in family 7.

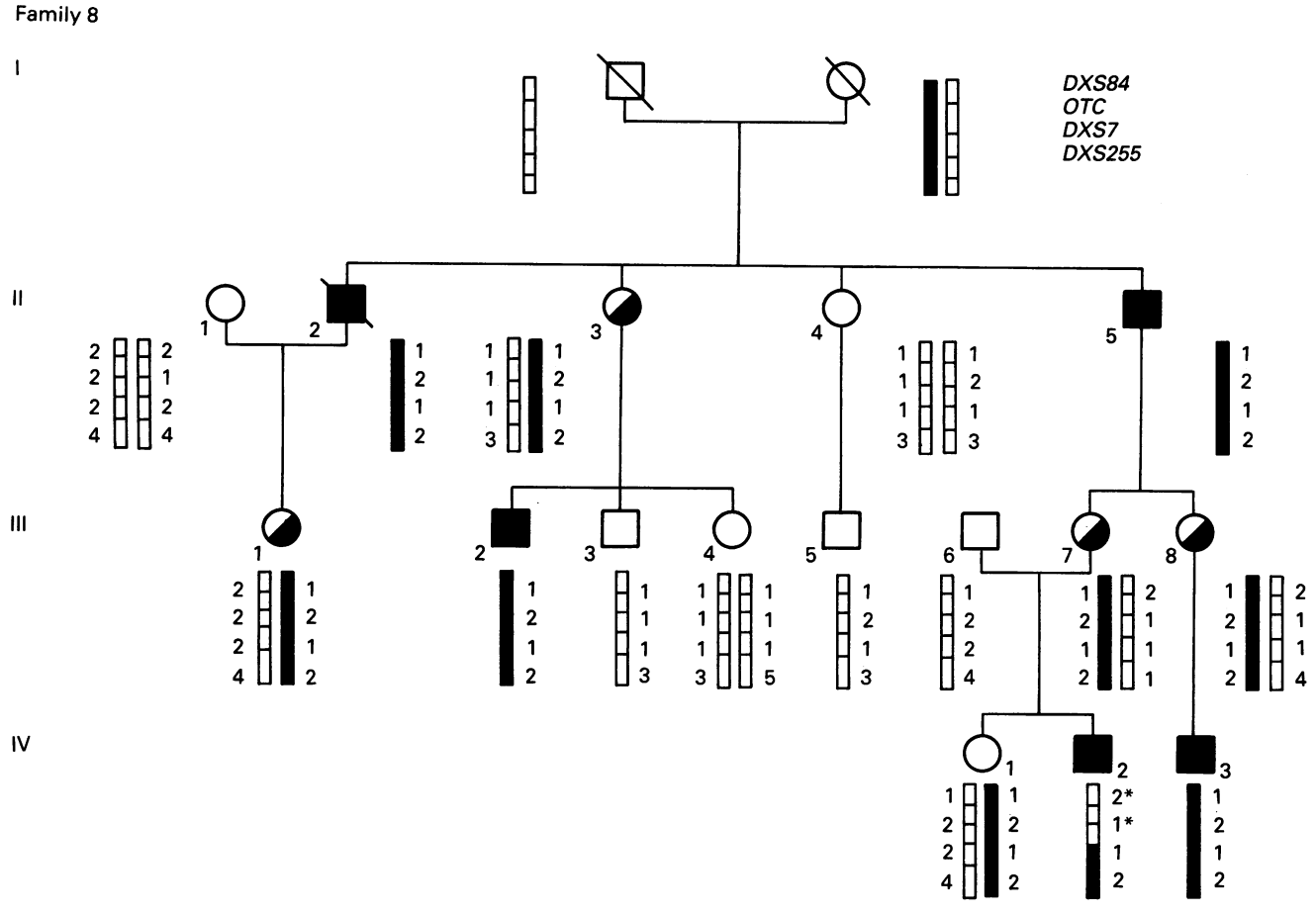


Figure 8 Segregation of RFLP alleles with XRLP in family 8. IV·2 was recombinant for DXS84 and OTC.

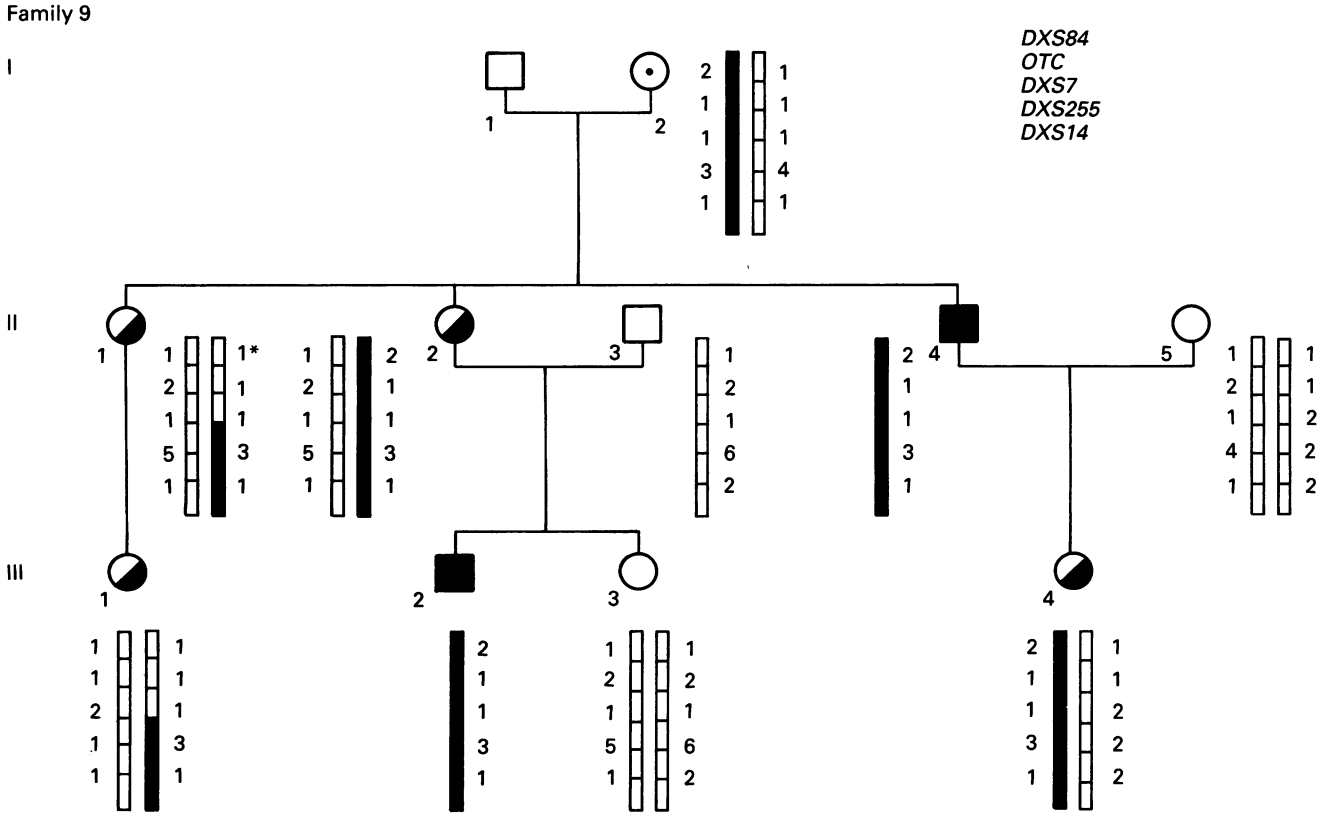


Figure 9 Segregation of RFLP alleles with XLRP in family 9.

Table 2 RFLP loci and probes on the short arm of the X chromosome used for analysis.

Locus	Probe	Location	Enzyme	Alleles	Frequencies
DXS84	754	Xp21.1	*Pst*I	12, 9	0·55, 0·45
OTC		Xp21.1	*Msp*I	6·6, 6·2	0·61, 0·39
				5·1, 4·4	0·73, 0·27
DXS7	L1.28	Xp11.3	*Taq*I	12, 9	0·77, 0·23
DXS255	M27β	Xp11.3-cen	*Pst*I	Multiallelic	
			*Msp*I	hypervariable	
			*Taq*I	systems	
DXS14	58.1	Xp11-cen	*Msp*I	4, 2·5	0·65, 0·35

Results

In XLRP families with night blindness as the onset symptom (XLRP-A), the maximum lod score was obtained with *OTC* ($\hat{Z}=4{\cdot}16$ at $\hat{\theta}=0$) while negative lod scores were obtained for *DXS255* and *DXS14* ($-4{\cdot}62$ and $-1{\cdot}91$ at $\theta=0{\cdot}01$ for *DXS255* and *DXS14* respectively) (table 3). By contrast, in XLRP families with early and severe myopia as the onset symptom (XLRP-B), the maximum lod score value was obtained at locus *DXS255* ($\hat{Z}=3{\cdot}13$ at $\hat{\theta}=0$) while negative values were obtained at loci *DXS84* and *OTC* ($Z=-2{\cdot}44$ and $Z=-0{\cdot}77$ at $\theta=0{\cdot}01$ for *DXS84* and *OTC* respectively) (table 4).

The location score method was used to estimate the position of the disease locus. In this procedure, the map of the marker loci is fixed and the position of the RP locus is allowed to vary throughout the map. The maximum likelihood estimate for location of the XLRP-A gene was close to the *OTC* locus (location score of 5·48 in log base 10 with no recombination events) while the maximum likelihood estimate for the XLRP-B gene was between *DXS7* and *DXS14* (maximum location score of 3·14 over *DXS255* with no recombination) (fig 10). Finally, genetic heterogeneity between XLRP-A and XLRP-B was shown at three genetic loci namely *DXS84*, *OTC*, and *DXS255*. Significant results were obtained with χ^2 values of 6·16 ($p<0{\cdot}05$), 17·75 ($p<0{\cdot}001$), and 12·88 ($p<0{\cdot}01$) for *DXS84*, *OTC*, and *DXS255* respectively. Finally, using the heterogeneity test HOMOG, conditional probabilities for each family being linked to the RP3 locus were 100% for families 1, 3, and 5 and 0 to 20% for families 8 and 9. Owing to the absence of recombination events no conclusion could be drawn for families 2, 4, 6, and 7. Consequently, the present study shows that XLRP with severe myopia as the onset symptom is accounted for by the RP2 gene while XLRP with night blindness as the onset symptom is accounted for by the RP3 gene.

Discussion

We have recently shown that different clinical profiles can be recognised in XLRP, according to age and mode of onset.[8] One clinical subtype, XLRP-B, has very early onset with severe myopia (mean age of onset (1 SD) = 3·5 (0·5) years). The other form, XLRP-A, starts later with night blindness sometimes associated with mild myopia (mean age of onset (1 SD) = 10·6 (4·1) years, $p=0{\cdot}01$[8]). Both forms meet the clinical criteria for RP according to the 1982 International Symposium of Ophthalmology[1] (see Patients and methods). Other diagnoses can be easily ruled out, especially X linked cone dystrophy (COD1) and X linked congenital stationary night blindness (CSNB1). Indeed, COD1 starts with a loss of visual acuity around 20 years of age, with frequent photophobia and dyschromatopsia. In this condition, the fundus is either normal or displays a tapetal-like sheen of the retina with no pigment deposits or narrowing of retinal vessels. This disease also differs from RP by the presence of a central scotoma and a marked abnormality in cone function with normal scotopic response at the start of the disease.[2] No confusion with CSNB1 can occur as this disease starts with congenital nystagmus, myopia, night blindness, and decreased vision with a normal looking fundus. By contrast with RP, visual function usually remains stationary throughout life and may vary among affected relatives.[27] Consequently, the clinical profiles reported here cannot be considered to be CSNB1 or COD1.

On the other hand, two gene loci have been recognised in XLRP. One disease gene is located on the proximal short arm of chromosome X (Xp11.21-p11.4), close to *DXS7* (*RP2*, linked to probe L1.28[9-11 14]). The other gene is distal to *RP2* and maps to Xp21.1, in the vicinity of the *OTC* locus (RP3[12 17 18]). In support of this, two cases of interstitial deletions in the Xp21.1 region have been reported in male patients with RP.[13 16] Previously, however, all studies have failed to distinguish any correlation between the clinical phenotype and the gene location.[17-19]

Table 3 Pairwise linkage results for XLRP-A with the five marker loci.

	0	0·01	0·05	0·1	0·2	0·3	0·4	$\hat{Z}$	$\hat{\theta}$
DXS84 (754)	$-\infty$	0·94	1·38	1·34	0·94	0·47	0·14	1·39	0·06
OTC	4·16	4·08	3·74	3·30	2·39	1·46	0·62	4·16	0
DXS7 (L1.28)	$-\infty$	0·19	0·72	0·81	0·70	0·48	0·23	0·81	0·11
DXS255 (M27β)	$-\infty$	$-4{\cdot}62$	$-1{\cdot}45$	$-0{\cdot}36$	0·28	0·30	0·15	0·33	0·25
DXS14 (58.1)	$-\infty$	$-1{\cdot}91$	$-0{\cdot}66$	$-0{\cdot}26$	$-0{\cdot}04$	$-0{\cdot}05$	$-0{\cdot}07$	0	0·50

Table 4 Pairwise linkage results for XLRP-B with the five marker loci.

	0	0·01	0·05	0·1	0·2	0·3	0·4	$\hat{Z}$	$\hat{\theta}$
DXS84 (754)	$-\infty$	$-2{\cdot}44$	$-1{\cdot}10$	$-0{\cdot}58$	$-0{\cdot}15$	0·009	0·05	0·05	0·39
OTC	$-\infty$	$-0{\cdot}77$	$-0{\cdot}10$	0·14	0·29	0·17	0·16	0·30	0·23
DXS7 (L1.28)	0·07	0·07	0·06	0·05	0·03	0·01	0·004	0·07	0
DXS255 (M27β)	3·13	3·08	2·84	2·52	1·87	1·18	0·53	3·13	0
DXS14 (58.1)	0·30	0·29	0·26	0·21	0·13	0·06	0·02	0·30	0

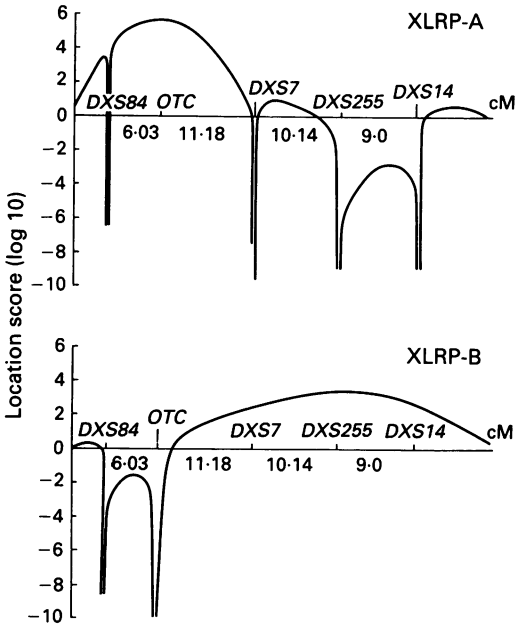


Figure 10 Support for possible locations of the RP gene with respect to the five marker loci in XLRP-A and XLRP-B families respectively. The location score method was used to estimate the position of the RP gene. Likelihood estimates are given in log base 10. Distances are shown between marker loci in centiMorgans along the map.

In the present study, we provide evidence for linkage of the two clinical subtypes with the RP2 and RP3 genes respectively. It appears, therefore, that the two markedly different modes of onset are accounted for by the two gene loci that have long been recognised on the basis of linkage studies. The form with early myopia as the onset symptom is accounted for by the RP2 locus while the form with later night blindness as the onset symptom is accounted for by the RP3 locus. Thus, the major difference between the two forms concerns the initial symptom, information which can be obtained from the parents and patients after careful questioning (especially as myopia may well occur in the course of all RP, regardless of the mode of inheritance[28]). By contrast, in adult life, no difference in either severity of the disease or aspect of the fundus was observed in our series regardless of the clinical subtype of XLRP. In fact, our XLRP-B probands were first myopic children but, when RP was diagnosed, the course was similar to that of XLRP-A patients whose disease started later. Both classes of patients displayed total night blindness, severe constriction of the visual field after 15 years of age, and markedly decreased visual acuity after 25 years.

Recently, Wright *et al*[29] reported a large kindred of X linked RP mapping to RP2. They emphasised that all affected males had myopia while all their unaffected sibs were emmetropic. They regarded this feature either as the pleiotropic effect of the RP2 gene or as a 'non-specific' symptom, related more to severity than to type. We also assume that a deprived pattern of vision early in life results in deprivation induced myopia whatever the cause. Along these lines, a significant degree of myopia was found in a retrospective study of refractive error among humans with various ocular anomalies disrupting the vision pattern.[30] In addition, it has been shown that restriction of only the peripheral field of vision in chicks led to the development of severe myopia.[31] In conclusion, the myopia discovered early in the first years of life in XLRP-B probands is likely to be related to the early onset of the disease which disrupts the normal process of emmetropisation.

As far as the status of heterozygotes is concerned, three different aspects of the retina have been reported in XLRP carriers, normal fundus, metallic sheen (tapetal-like reflex), or peripheral pigmentary deposits.[4 6 32-34] In our series, however, no correlation could be found between one particular aspect of the fundus and either clinical subtype of the disease. Musarella *et al*[18] failed to establish any relationship between the genotype and the retinal phenotype of carriers, and ERG was not found to be discriminative either.[35 36]

In conclusion, this study emphasises the primary importance of questioning patients and families carefully about the early history of the disease, since detecting phenotypic differences at an advanced stage of the illness may be very difficult. This does not hold for RP only, but for other genetic disorders as well, especially when more than one gene is thought to account for one single clinical phenotype (for example, Marfan syndrome and tuberous sclerosis[37-39]).

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1 Marmor MF, Aguirre G, Arden G, Berson E, Birch DG. Retinitis pigmentosa, a symposium on terminology and methods of examination. *Ophthalmology* 1983;**90**:126–31.
2 Pagon RA. Retinitis pigmentosa. *Surv Ophthalmol* 1988;**33**:137–77.
3 Berson EL, Rosner B, Simonoff E. Risk factors for genetic typing and detection in retinitis pigmentosa. *Am J Ophthalmol* 1980;**89**:763–75.
4 Boughman JA, Conneally PM, Nance WE. Population genetic studies of retinitis pigmentosa. *Am J Hum Genet* 1980;**32**:223–35.
5 Jay M. On the heredity of retinitis pigmentosa. *Br J Ophthalmol* 1982;**66**:405–16.
6 Bundey S, Crews SJ. A study of retinitis pigmentosa in the city of Birmingham. II. Clinical genetic heterogeneity. *J Med Genet* 1984;**21**:421–8.
7 Humphries P, Farrar GJ, Kenna P, McWilliam P. Retinitis pigmentosa: genetic mapping in X-linked and autosomal forms of the disease. *Clin Genet* 1990;**38**:1–13.
8 Kaplan J, Bonneau D, Frézal J, Munnich A, Dufier JL. Clinical and genetic heterogeneity in retinitis pigmentosa. *Hum Genet* 1990;**85**:635–42.
9 Bhattacharya SS, Wright AF, Clayton JF, *et al*. Close genetic linkage between X-linked retinitis pigmentosa and a restriction fragment length polymorphism identified by recombinant DNA probe L1.28. *Nature* 1984;**309**:253–6.
10 Friedrich U, Warburg M, Wieacker P, Wienker TF, Gal A, Ropers HH. X-linked retinitis pigmentosa: linkage with the centromere and a cloned DNA sequence from the proximal short arm of the X chromosome. *Hum Genet* 1985;**71**:93–9.
11 Mukai S, Dryja TP, Bruns GAP, Aldridge JF, Berson EL. Linkage between the X-linked retinitis pigmentosa locus and the L1.28 locus. *Am J Ophthalmol* 1985;**100**:225–9.
12 Nussbaum RL, Lewis RA, Lesko JG, Ferrel R. Mapping ophthalmological disease. II. Linkage of relationship of

X-linked retinitis pigmentosa to X chromosome short arm markers. *Hum Genet* 1985;**70**:45–50.
13 Francke U, Ochs HD, de Martinville B, *et al.* Minor Xp21 chromosome deletion in a male associated with expression of Duchenne muscular dystrophy, chronic granulomatous disease, retinitis pigmentosa and McLeod syndrome. *Am J Hum Genet* 1985;**37**:250–67.
14 Wright AF, Bhattacharya SS, Clayton JF, *et al.* Linkage relationships between X-linked retinitis pigmentosa and nine short arm markers: exclusion of the disease locus from Xp21 and localization to between DXS7 and DXS14. *Am J Hum Genet* 1987;**41**:635–44.
15 Wirth B, Denton MJ, Chen JD, *et al.* Two different genes for X-linked retinitis pigmentosa. *Genomics* 1988;**2**:263–6.
16 De Saint-Basile G, Bohler MC, Fischer A, *et al.* Xp21 DNA microdeletion in a patient with chronic granulomatous disease, retinitis pigmentosa, and the McLeod phenotype. *Hum Genet* 1988;**80**:85–9.
17 Chen JD, Dickinson P, Gray R, Constable I, Sheffield L, Denton MJ. Non-allelic mutations in X-linked retinitis pigmentosa. *Clin Genet* 1989;**35**:338–42.
18 Musarella MA, Burghes A, Anson-Cartwright L, *et al.* Localization of the gene for X-linked recessive type of retinitis pigmentosa (XLRP) to Xp21 by linkage analysis. *Am J Hum Genet* 1988;**43**:484–94.
19 Musarella MA, Anson-Cartwright L, Leal SM, *et al.* Multipoint linkage analysis and heterogeneity testing in 20 X-linked retinitis pigmentosa families. *Genomics* 1990;**8**:286–96.
20 Rigby PWJ, Dieckmann M, Rhodes C, Berg P. Labelling deoxyribonucleic acid to high specific activity in vitro by nick translation with DNA polymerase I. *J Mol Biol* 1977;**113**:237–51.
21 Lathrop GM, Lalouel M. Easy calculation of lod-scores and genetic risks on small computers. *Am J Hum Genet* 1984;**36**:460–5.
22 Lathrop GM, Lalouel JM, Julier C, Ott J. Multilocus linkage analysis in humans: detection of linkage and estimation of recombination. *Am J Hum Genet* 1985;**37**:482–98.
23 Drayna D, White R. The genetic linkage map of the X chromosome. *Science* 1985;**230**:753–8.
24 Goodfellow PN, Davies KE, Ropers HH. Report of the committee on genetic constitution of the X and Y chromosomes. HGM8. *Cytogenet Cell Genet* 1985;**40**:296–352.
25 Mahtani ME, Willard HF. A primary genetic map of the pericentric region of the human X chromosome. *Genomics* 1988;**2**:294–301.
26 Ott J. Variability of the recombination fraction. Analysis of human genetic linkage. Baltimore: Johns Hopkins University Press, 1985:112–5.
27 Lambert SR, Taylor D, Kriss A. The infant with nystagmus, normal appearing fundi, but an abnormal ERG. *Surv Ophthalmol* 1989;**34**:173–86.
28 Sieving PA, Fishman GA. Refractive errors of retinitis pigmentosa patients. *Br J Ophthalmol* 1978;**62**:163–7.
29 Wright AF, Bhattacharya SS, Aldred MA, *et al.* Genetic localisation of the RP2 type of X-linked retinitis pigmentosa in a large kindred. *J Med Genet* 1991;**28**:453–7.
30 Rabin J, Van Sluyters RC, Malach R. Emmetropization: a vision dependent phenomenon. *Invest Ophthalmol Visual Sci* 1981;**20**:561–4.
31 Wallman J, Turkel J, Trachtman J. Extreme myopia produced by modest change in early visual experience. *Science* 1978;**20**:1249.
32 Bird AC. X-linked retinis pigmentosa. *Br J Ophthalmol* 1975;**59**:177–99.
33 Fishman GA, Weinberg AW, McMahon TT. X-linked recessive retinitis pigmentosa: clinical characteristics of carriers. *Arch Ophthalmol* 1986;**104**:1329–35.
34 Musarella MA, Anson-Cartwright L, Burghes A, Worton RG, Lesko JG, Nussbaum RL. Linkage analysis of a large Latin-American family with X-linked retinitis pigmentosa and metallic sheen in the heterozygote carrier. *Genomics* 1989;**4**:601–5.
35 Arden GB, Carter RM, Hogg CR. A modified ERG technique and the results obtained in X-linked retinitis pigmentosa. *Br J Ophthalmol* 1983;**67**:419–30.
36 Berson EL, Rosen JB, Simonoff EA. Electroretinographic testing as an aid in detection of carriers of X-chromosome-linked retinitis pigmentosa. *Am J Ophthalmol* 1979;**87**:460–8.
37 Boileau C, Jondeau G, Coulon M, *et al.* Linkage studies for Marfan syndrome and for markers on chromosomes 13 and 15. *Abstracts Am Soc Hum Genet, Cincinnati, USA, 16–20 October 1990.*
38 Sampson JR, Yates JRW, Pirrit LA, *et al.* Evidence for genetic heterogeneity in tuberous sclerosis. *J Med Genet* 1989;**26**:511–6.
39 Janssen LAJ, Sandkuyl LA, Merkens EC, *et al.* Genetic heterogeneity in tuberous sclerosis. *Genomics* 1990;**8**:237–42.