Genetic heterogeneity of primary open angle glaucoma and ocular hypertension: linkage to GLC1A associated with an increased risk of severe glaucomatous optic neuropathy

Antoine P Brézin, Alain Béchetoille, Pascale Hamard, Françoise Valtot, Merouane Berkani, Ahmed Belmouden, Marie F Adam, Stéphane Dupont de Dinechin, Jean-François Bach, Henri-Jean Garchon

Abstract

The GLC1A locus for autosomal dominant juvenile and middle age onset primary open angle glaucoma (OAG) has been mapped to chromosome 1q21-q31. OAG, however, is a heterogeneous disease. We tested linkage of OAG and ocular hypertension (OHT), a major risk factor for OAG, to GLC1A in eight French families with multiple cases of juvenile and middle age onset OAG. There was strong evidence of genetic heterogeneity, four families being linked to GLC1A and two or three others being unlinked, depending on whether the complete OAG phenotype was analysed alone or jointly with OHT. Peak intraocular pressure (IOP) did not differ significantly between the two groups of families, while linkage to GLC1A conferred a highly increased risk of developing OAG and of having severe glaucomatous optic neuropathy. Testing linkage of familial OAG to GLC1A may therefore have prognostic value too.

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Glaucoma is the second most common cause of irreversible visual impairment in industrialised countries, with an estimated worldwide prevalence of between 1 and 2%.¹ Primary open angle glaucoma (OAG) combines a particular type of damage to the optic disc with a progressive loss of visual field.^{2,3} Glaucomatous optic atrophy is primarily characterised by an excavated appearance of the disc. This definition of glaucoma, however, encompasses heterogeneous phenotypes.⁴ Ocular hypertension (OHT) is not a diagnostic criterion for glaucoma, but occurs in a large proportion of patients. It is the most consistent risk factor for OAG and can precede the development of glaucomatous optic neuropathy. In animal models, laser induced OHT results in optic disc cupping.⁵ Lowering intraocular pressure (IOP) is currently the best treatment for glaucoma.⁶ Taken together, these observations suggest that, at least in certain forms of glaucoma, raised IOP is a pathogenic mechanism in the development of OAG in humans.

The precise pathogenesis of OHT and OAG is unclear. Epidemiological studies have emphasised the importance of a family history of OAG as a risk factor, strongly suggesting that OAG is influenced by genetic determinants.¹ The juvenile form of OAG (diagnosed before the age of 40 years) is often inherited in a simple mendelian manner, with a highly penetrant autosomal mode of transmission.⁷ Clinically, juvenile onset OAG comprises particularly raised IOP (often above 35 mm Hg) resulting in severe optic disc cupping. Linkage analysis of a family affected by juvenile OAG has mapped the disease causing gene to chromosome 1q21-q31,^{8,9} currently within a 3 cM interval.^{10,11}

In initial linkage studies, disease onset consistently occurred before the age of 20.^{9,12-15} Subsequently, two large families were characterised, in which both juvenile OAG and middle age onset OAG were observed.^{16,17} These findings indicated that the range of phenotypes determined by the GLC1A chromosomal region could be broader than initially thought. However, GLC1A was also probably excluded in one family with middle age onset OAG,¹⁸ and a second localisation (GLC1B) on proximal chromosome 2q was recently proposed.¹⁹ Therefore, these results raised questions as to the role of the GLC1A locus in middle to late age onset OAG and OHT, which are much more prevalent than juvenile OAG in the general population. To address this essential issue, we characterised eight families with two or more cases of OAG associated with OHT. We then assessed linkage of OAG or OHT or both to GLC1A.

Materials and methods

Information on the possibility of testing linkage to the GLC1A locus in families with five or more members affected by OAG or OHT was communicated to French ophthalmologists. Eight consecutive families meeting this requirement and comprising 177 living or dead subjects were included in the study. Informed consent and blood samples were obtained from 137 of the 145 living subjects, in keeping with French legislation.

The complete ophthalmological records of each subject were requested from local ophthalmologists. Clinical assessment included slit

INSERM U25, Hôpital Necker, 161 rue de Sévres, 75743 Paris Cedex 15, France
A P Brézin
A Belmouden
M F Adam
S Dupont de Dinechin
J-F Bach
H-J Garchon

Service d'Ophthalmologie, CHU Angers, Angers, France
A Béchetoille

CHNO Quinze-vingts, Paris, France
P Hamard
F Valtot

Institut du Glaucome, Hôpital Saint-Joseph, Paris, France
F Valtot
M Berkani

Correspondence to: Dr Garchon.

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lamp biomicroscopy, applanation tonometry, gonioscopy, and visual field testing, using automated perimeter (Humphrey field analyser, Octopus™, or Moniteur Ophtalmologique™). Ophthalmological examination was performed by at least one of us (APB, AB, PH, FV, or MB) on 128 subjects (93% of living subjects). For the purposes of genetic linkage analysis, OAG was diagnosed by the combination of open angle gonioscopy and characteristic cupping of the optic disc with a loss of visual field. Isolated OHT was defined by an IOP ≥ 22 mm Hg on at least two occasions with a normal appearance of the optic disc and a normal visual field. The IOP threshold of 22 mm Hg was retained because it is over two standard deviations above the mean¹ and it is associated with a dramatic increase in the risk of developing glaucoma.^{20 21}

Genomic DNA was extracted from purified white blood cells, using established procedures.²² Microsatellite markers spanning the GLC1A susceptibility region were chosen from the Génethon linkage map.²³ The NGA1 tetranucleotide repeat was characterised in our laboratory.¹¹ Oligonucleotides were purchased from Genosys™ with one fluorescently labelled oligonucleotide among each pair of primers (6-FAM for D1S194, D1S196, D1S445, D1S433, D1S452, D1S210, and D1S218, JOE for D1S2851 and NGA1, and TAMRA for D1S242). A fourth dye was used for size standards (ROX 350, Applied Biosystems™). Up to six amplification products were coprecipitated and comigrated in a 6% polyacrylamide denaturing gel on an Applied Biosystem 373 Sequencer. The molecular weight of amplified products was calculated by using GENESCAN 1.0 and GENOTYPER 1.0 software.

Pedigree data were exported for linkage analysis with FASTLINK software version 3.0.^{24 25} Because little is known of the actual frequency of the GLC1A gene in the general population, and also because disease parameters may be critical when testing a model,²⁶ four sets of disease parameters were used to test linkage to GLC1A. They were deduced from four values of the proportion of genetic cases (cases caused by the GLC1A locus). These four values (0.01, 0.03, 0.1, and 0.3) were estimated from the published figures of OAG prevalence at various ages¹ and were expected broadly to cover the actual value. A prevalence rate of 1% in the general population was selected. In all cases, a dominant mode of transmission and a penetrance rate of 0.9 for the disease gene were assumed. The disease gene frequency P was calculated from the frequency of genetically affected subjects (A) using the formula:

$$A = P^2 F_{DD} + 2P(1-P)F_{Dd}$$

where F_{DD} and F_{Dd} are the penetrance rates of the disease gene in the homozygous and heterozygous state, respectively, whereas the frequency of phenocopies was given by:

$$C = (1-P)^2 F_{dd}$$

where F_{dd} is the penetrance of the wild type allele. The overall disease frequency (prevalence) is $Prev = A + C$. For clarity, only data

obtained from 30% of genetic cases are presented here.

Multipoint analysis was done with the VITESSE algorithm.²⁷ Allele frequencies of microsatellite markers were determined in a cohort of 96 spouses from our family set. Family admixture was tested with the HOMOG program.²³

Clinical features were compared between GLC1A linked and unlinked families by assuming a logistic regression model, taking GLC1A linkage as the dependent variable. Exact estimates of the odds ratios (OR) and their two sided p values were obtained with LogXact Turbo software.²⁸

Results

CLINICAL FEATURES

We studied 137 living subjects from eight multigeneration families (fig 1). Affected members were found in two to five generations of each family. Of 60 affected patients, 31 were men and 29 were women. Forty-five patients had definite OAG and 15 isolated OHT (table 1). Glaucomatous neuropathy was observed in at least two subjects in each family. There was no evidence of secondary glaucoma, and gonioscopy was normal in all eyes. The age at diagnosis (OHT or OAG) ranged from 7 to 82 years, with a median of 28.5 years (28 years for patients with OAG). A marked rise of IOP was found in some or all affected members of each family, 30 patients having recorded IOP values of 30 mm Hg or more in at least one eye. Visual field testing showed an advanced scotoma in at least one eye in 24 patients. Glaucomatous optic atrophy was responsible for a total loss of light perception in 12 eyes. Advanced cupping of the optic disc, with a c/d ratio of 0.9 or more, was observed in 34 eyes. Thirty-four patients had undergone surgery.

Although the clinical features of the probands were similar in all eight families, there seemed to be two subgroups of families with regard to the frequency of isolated OHT, which was not observed in families A-D, and of severe glaucomatous optic neuropathy, which seemed to be more frequent in these four families. These differences are analysed further below in the light of linkage data.

LINKAGE ANALYSIS AND GENETIC HETEROGENEITY

Segregation analysis of OHT and OAG indicated that more than 50% of children of affected parents were affected, and that there was no difference in the rate of paternal versus maternal transmission of disease. There was one obligate carrier in family G. Another carrier (II.2) died at the age of 34, before his clinical status could be documented. A disease model with an autosomal dominant mode of transmission and a penetrance rate of 90% was therefore adopted to test linkage of OAG/OHT to GLC1A in these eight families. Because the proportion of cases of glaucoma caused by GLC1A, that is, the proportion of genetic cases, is largely unknown, and because its value may markedly alter the significance of the results, four values (1% to 30%) were

Table 1 Clinical characteristics of affected family members

Patient	Age	Diagnosis	Age at diagnosis	Highest IOP (either eye)	c/d ratio		Visual field		Surgical treatment (either eye)
					OD	OS	OD	OS	
Family A									
III.1	82	OAG	31	35	1.0	1.0	↓↓↓↓	↓↓↓↓	Yes
III.3	75	OAG	37	35	1.0	1.0	↓↓↓	↓↓↓	Yes
III.4	73	OAG	40	40	0.9	0.9	↓↓↓	↓↓↓	Yes
IV.2	51	OAG	36	34	*	*	↓↓↓	↓↓↓	Yes
IV.6	52	OAG	30	23	0.3	0.3	↓↓↓	↓↓	Yes
V.1	26	OAG	22	26	0.7	0.8	↓↓	↓↓	No
V.2	24	OAG	20	24	0.2	0.2	↓↓	↓↓	No
Family B									
I.1	93	OAG	20	NA	1.0	1.0	↓↓↓↓	↓↓↓↓	No
II.2	70	OAG	28	NA	1.0	1.0	↓↓↓↓	↓↓↓↓	Yes
II.3	49	OAG	28	25	0.6	0.8	↓	↓	Yes
III.2	45	OAG	25	40	0.8	0.9	↓↓↓	↓↓↓	Yes
III.3	36	OAG	18	35	0.9	1.0	↓↓↓	↓↓↓↓	Yes
IV.1	17	OAG	11	32	0.2	0.2	↓	↓	No
IV.2	20	OAG	18	27	0.3	0.4	↓	↓	No
Family C									
II.2	71	OAG	29	37	1.0	0.9	↓↓↓	↓↓↓	Yes
II.3	73	OAG	46	28	1.0	0.9	↓↓↓	↓↓↓	Yes
II.4	74	OAG	51	48	NV	NV	↓↓↓	↓↓↓	Yes
III.2	47	OAG	40	35	0.9	0.9	↓↓↓	↓↓↓	Yes
IV.1	24	OAG	23	40	0.5	0.6	↓↓	↓↓	No
IV.2	22	OAG	21	28	0.9	0.9	↓↓↓	↓↓	Yes
Family D									
III.2	74	OAG	27	NA	1.0	1.0	↓↓↓	↓↓↓	Yes
IV.2	44	OAG	8	NA	0.8	0.8	↓↓↓	↓↓↓	Yes
IV.4	53	OAG	11	NA	1.0	1.0	↓↓↓	↓↓↓↓	Yes
IV.6	49	OAG	12	40	1.0	1.0	↓↓↓	↓↓↓	Yes
IV.9	48	OAG	11	45	0.8	0.6	↓↓	↓↓	Yes
IV.11	51	OAG	17	NA	NV	NV	↓↓	↓	Yes
V.1	9	OAG	7	36	0.7	0.7	↓↓	↓↓	Yes
V.4	25	OAG	8	40	0.9	1.0	↓↓↓	↓↓↓	Yes
V.6	25	OAG	10	35	0.4	0.3	NA	NA	Yes
V.8	28	OAG	12	40	0.7	0.7	↓↓↓	↓↓	Yes
Family E									
II.2	64	OAG	42	40	0.5	†	↓	†	Yes
II.3	70	OAG	40	51	NV	NV	NA	NA	Yes
III.2	32	OAG	19	24	0.3	0.3	↓	↓	No
III.3	34	OHT	16	35	0.3	0.3	N	N	No
III.5	38	OHT	28	30	0.1	0.1‡	N	‡	No
III.7	44	OAG	19	45	NV	NV	↓↓↓	↓↓↓↓	Yes
III.9	53	OAG	32	45	0.6	0.2	↓↓	↓	Yes
Family F									
I.2	87	OAG	82	58	1.0	1.0	↓↓↓↓	↓↓↓↓	Yes
II.2	57	OHT	54	22	0.1	0.2	N	N	No
II.4	46	OHT	43	24	0.4	0.3	N	N	No
II.6	59	OAG	56	25	0.8	0.8	↓	↓	Yes
II.7	62	OAG	44	NA	1.0	1.0	↓↓↓↓	↓↓↓	Yes
III.4	21	OHT	19	23	0.2	0.1	N	N	No
III.5	38	OHT	33	28	0.4	0.3	N	N	No
III.7	35	OHT	32	27	0.5	0.4	N	N	No
III.8	34	OHT	32	22	0.3	0.3	N	N	No
III.9	33	OAG	31	28	0.8	0.8	↓	↓	No
III.10	32	OHT	30	24	0.8	0.7	N	N	No
III.11	31	OHT	25	22	0.2	0.3	N	N	No
Family G									
IV.2	65	OAG	60	38	0.4	0.3	N	↓	No
IV.3	63	OAG	58	25	0.5	0.6	↓	↓	No
IV.5	51	OAG	47	32	0.6	NV	↓	↓	Yes
IV.8	51	OHT	34	24	0.2	0.2	N	N	No
IV.10	59	OHT	50	28	0.5	0.6	N	N	No
V.1	41	OAG	36	34	0.9	0.8	↓↓↓	↓↓	Yes
Family H									
II.2	39	OAG	38	37	0.5	0.6	N	↓	Yes
II.3	34	OAG	34	36	0.5	0.4	↓	↓	No
III.1	18	OHT	17	27	0.2	0.2	N	N	No
III.5	17	OHT	16	33	0.3	0.3	N	N	No
III.6	14	OHT	13	28	0.2	0.3	N	N	No

IOP is in mm Hg. NA: not available, NV: not visualised. * Tilted disc syndrome. † Trauma. ‡ Unilateral myopia. Visual field: N = normal, ↓ = early defect, ↓↓ = arcuate scotoma or nasal step, ↓↓↓ = advanced scotoma, ↓↓↓↓ = no light perception.

considered on the basis of epidemiological data (see Materials and methods). Pairwise lod score data obtained with an assumed proportion of genetic cases of 30% are shown for five of the eight markers tested in table 2 (data not shown for D1S210, D1S452, and D1S242). Four families (A-D) had a positive lod score with two or more markers. Family D had a lod score above 3 with five of the eight markers tested across the susceptibility region. Conversely, three other families (E, F, H) had a lod score <−2 with three or more markers,

suggesting an absence of linkage. When the proportion of genetic cases was reduced, positive lod scores (families A-D) hardly varied, whereas negative lod scores fell by one unit or more, therefore increasing evidence against linkage (data not shown). To test further the heterogeneity of linkage, four point analysis was carried out across two successive and overlapping subsets of markers (D1S194-D1S445-D1S2851 and D1S445-D1S2851-D1S218) encompassing the GLC1A interval. Testing family admixture with the

interval.¹¹ No conclusion could be drawn in the case of family G (posterior probability of being linked 0.25, 95% confidence interval 0.04–0.71). As for family H, the D1S445 marker not being informative in this pedigree, two additional markers closely linked to D1S445, D1S196 and D1S433, were typed in this family and six point analysis was conducted with the D1S194–D1S196–D1S433–D1S2851–D1S218 grid of markers. This analysis clearly excluded linkage of the OAG/OHT to GLC1A in pedigree H (fig 2B), with lod scores <–2 from D1S196 to and beyond D1S218.

Because OHT is not a perfect predictor of OAG, linkage analysis including only subjects affected with OAG was repeated in families E–H. Despite a smaller number of informative meioses, linkage of OAG alone to GLC1A was excluded by a four point analysis in families E

Table 2 Two point lod scores for OAG/OHT and GLC1A locus microsatellite markers in eight families

Markers	θ					Zmax	θ (%)
	0.0	0.01	0.05	0.1	0.2		
D1S194							
A	-0.21	0.12	0.51	0.62	0.58	0.63	0.126
B	1.62	1.60	1.49	1.36	1.07	1.62	0.001
C	0.52	0.50	0.44	0.36	0.21	0.52	0.001
D	1.17	1.14	1.02	0.87	0.59	1.17	0.001
E	-2.24	-1.08	-0.46	-0.22	-0.04		> 0.5
F	-1.55	-0.32	-0.86	-0.58	-0.27		> 0.5
G	0.38	0.45	0.57	0.59	0.46	0.59	0.082
H	-0.57	-0.24	0.18	0.33	0.38	0.38	0.173
D1S445							
A	0.23	0.26	0.35	0.37	0.33	0.37	0.105
B	0.59	0.58	0.53	0.46	0.33	0.59	0.001
C	-0.16	-0.15	-0.12	-0.09	-0.05		> 0.5
D	0.46	0.91	1.29	1.33	1.11	1.33	0.085
E	-0.95	-0.71	-0.33	-0.14	-0.01		> 0.5
F	-2.91	-2.80	-2.12	-1.45	-0.58	0.05	0.428
G	-1.20	-1.10	-0.63	-0.27	0.03	0.96	0.301
H	0.10	0.10	0.09	0.07	0.04	0.10	0.001
D1S2851							
A	1.46	1.48	1.51	1.45	1.18	1.51	0.041
B	1.41	1.39	1.28	1.15	0.88	1.41	0.001
C	1.31	0.29	1.17	1.02	0.72	1.31	0.001
D	3.92	3.85	3.58	3.22	2.46	3.92	0.001
E	-1.77	-0.55	0.05	0.23	0.29	0.30	0.182
F	-1.74	-1.14	-0.25	0.14	0.41	0.43	0.248
G	-1.72	-1.44	-1.07	-0.89	-0.60		> 0.5
H	-3.10	-1.52	-0.43	0.00	0.30	0.34	0.265
NGA1							
A	0.49	0.60	0.78	0.82	0.69	0.82	0.090
B	2.01	1.97	1.83	1.66	1.28	2.01	0.001
C	1.24	1.21	1.10	0.96	0.67	1.24	0.001
D	3.37	3.30	3.04	2.71	2.03	3.37	0.001
E	-3.45	-2.10	-0.98	-0.48	-0.08		> 0.5
F	-2.28	-2.27	-2.06	-1.56	-0.77		> 0.5
G	-1.23	-1.06	-0.64	-0.38	-0.16		> 0.5
H	-3.34	-1.77	-0.67	-0.21	0.13	0.21	0.301
D1S218							
A	0.75	0.82	0.96	1.02	0.92	1.02	0.103
B	0.71	0.73	0.76	0.77	0.69	0.77	0.075
C	1.32	1.29	1.17	1.03	0.73	1.32	0.001
D	2.16	2.12	1.94	1.71	1.25	2.16	0.001
E	-3.09	-1.75	-0.89	-0.48	-0.12		> 0.5
F	-5.51	-3.91	-1.81	-0.83	-0.03	0.24	0.340
G	-0.92	-0.64	-0.28	-0.14	0.02		> 0.5
H	-3.12	-1.52	-0.43	0.00	0.30	0.34	0.264

and F, throughout the D1S2851-D1S218 interval (fig 2C). No conclusion could be drawn for families G and H (not shown).

Altogether linkage analysis provided strong evidence of genetic heterogeneity. Linkage to GLC1A was supported in four families (A-D) and was excluded in two or three other families (E, F or E, F, H), depending on whether the complete OAG phenotype was analysed alone or jointly with OHT.

Table 3 Comparison of phenotypes in GLC1A linked and GLC1A unlinked families

	Odds ratio (95% confidence interval)	p value
Exclusion based on OAG and OHT cases		
Age at diagnosis	0.95 (0.90-0.99)	0.011
Age at diagnosis, excluding family D	0.97 (0.93-1.02)	NS
Peak IOP	1.04 (0.99-1.09)	NS
Cup/disc ratio	49.9 (5.36-634)	0.0002
Advanced scotoma in at least one eye	3.35 (1.88-6.38)	< 10 ⁻⁴
Filtration surgery in at least one eye	7.64 (2.03-33)	0.0013
Isolated OHT	44.2 (6.58-∞)	< 10 ⁻⁴
Exclusion based on OAG cases only		
Age at diagnosis	0.93 (0.88-0.98)	0.003
Age at diagnosis, excluding family D	0.96 (0.90-1.01)	NS
Peak IOP	1.03 (0.99-1.09)	NS
Cup/disc ratio	30.3 (3.13-390)	0.002
Advanced scotoma in at least one eye	2.97 (1.66-6.10)	< 10 ⁻⁴
Filtration surgery in at least one eye	6.54 (1.6-30.6)	0.006
Isolated OHT	40.3 (5.7-∞)	< 10 ⁻⁴

NS = not significant.

GENETIC HETEROGENEITY CORRELATES WITH PHENOTYPIC HETEROGENEITY

A logistic regression model was used to compare phenotypes in GLC1A linked (A-D) and unlinked families. This comparison lent strong statistical support to the feeling that two subgroups of families could be identified on the basis of disease frequency and severity in each family. Whether linkage was excluded on the basis of both OAG and OHT or OAG alone, similar conclusions could be reached (table 3). Linkage to GLC1A conferred a highly increased risk of glaucoma as opposed to isolated OHT (odds ratio, OR=44.2, p<10⁻⁴; OR=40.3, p<10⁻⁴, respectively), and of severe glaucomatous optic neuropathy, as judged by optic disc cupping (OR=49.9, p=0.0002; OR=30.3, p=0.002), severe impairment of visual field (OR=3.35, p<10⁻⁴; OR=2.97, p<10⁻⁴), and indications for surgery (OR=7.64, p=0.0013; OR=6.54, p=0.006). In striking contrast, peak IOP was not different between the two sets of families, even after adjusting for confounding factors such as family size and age at the time of diagnosis.

Age at the time of diagnosis was mildly influenced by linkage to GLC1A and was slightly younger in GLC1A linked than in GLC1A unlinked families (OR=0.95,

Figure 2 Multipoint analysis of OAG/OHT (A, B) or OAG only (C) for families E, F (A, C), and H (B). The grid of test markers is indicated above each plot. Distances (x axis) are in cM. They were drawn from the Gnthon linkage map²³ or were deduced from the physical map of the GLC1A region, in which 1 cM is approximately equivalent to 1 Mb.¹¹ Four sets of disease parameters were used (see Materials and methods). Only results corresponding to a proportion of 0.3 genetic cases are shown. Other proportions tested (0.1, 0.03, and 0.01) yielded smaller multipoint lod scores, therefore increasing evidence against linkage in these families.

$p=0.011$; $OR=0.93$, $p=0.003$). However, the group of GLC1A linked families by itself was heterogeneous regarding this criterion, as age at the time of diagnosis was significantly younger in family D than in the other three GLC1A linked families ($OR=1.31$, 95% confidence interval=1.11–1.66, $p<10^{-4}$). The differ-

ence between the two sets of families disappeared after removal of family D.

Discussion

Raised IOP, as a main risk factor for OAG, is likely to play a major role in its pathophysiology. Taking OHT into consideration was therefore legitimate and usefully increased the number of informative meioses, notably in families E–H. Indeed, the segregation pattern of OHT/OAG in these families was compatible with autosomal dominant inheritance and high penetrance. In this context, joint linkage analysis of OAG and OHT showed heterogeneity with regard to linkage to GLC1A. Linkage in the specified model and using a broad range of disease parameters was unambiguously ruled out in three families, whereas no conclusion could be drawn regarding a fourth family. However, because the relationship between OHT and OAG is not unequivocal, it was critical also to restrict our analysis to OAG only cases. Importantly, in two of the three unlinked families, linkage to GLC1A remained excluded.

An unknown number of genes not located on chromosome 1q may play a role in these GLC1A unlinked families. Other loci for rare forms of glaucoma, including glaucoma associated with iris hypoplasia²⁹ and congenital glaucoma,³⁰ were recently mapped to chromosomes 4q and 2p, respectively. These regions are unlikely candidates for linkage with OAG and OHT in our GLC1A unlinked families, as their clinical features differ markedly from those of both recently mapped forms of glaucoma. More relevant to our discussion is the recent localisation of a second locus for OAG, GLC1B, on chromosome 2qcen–q13.¹⁹ However, none of our families (E–H) appeared to be linked to GLC1B, whether OAG only or OAG/OHT cases were included in the linkage analysis (MFA and HJG, unpublished data), suggesting the existence of at least a third disease locus.

The most remarkable finding in this study resulted from the comparison of clinical features in GLC1A linked and unlinked families. Linkage to GLC1A appeared to be a major risk factor for the development of glaucoma and severe glaucomatous optic neuropathy, as judged by various criteria including optic disc cupping, severe alteration of the visual field, and the decision to undertake surgical treatment, whereas it did not influence the magnitude of the increase in IOP. In the previously reported GLC1A unlinked family, severity of disease varied greatly. Therefore, lack of linkage to GLC1A would predict a better prognosis. In a recent 12 year follow up study of persons with OHT and an initially normal visual field by Quigley *et al*,³¹ surprisingly, a family history of glaucoma was not a significant risk factor for OAG in a subject with isolated OHT. Risk factors identified in this work and relevant to our findings included older age, higher IOP, characteristic modifications of the optic disc, and alterations of the nerve fibre layer at the initial visit. The latter two risk factors are in fact early signs of optic

nerve damage. It would be most interesting to test whether a family history of glaucoma with GLC1A linkage is also a significant risk factor for developing glaucoma in such a cohort of subjects with isolated OHT. Our results should also have a significant role for optimising the management of members of glaucoma families, as raised IOP is probably a reliable parameter of disease evolution in GLC1A linked families, whereas it may be less valuable in GLC1A unlinked families.

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