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Adult-onset primary glaucoma and molecular genetics: A review

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PURPOSE. *To evaluate recent molecular genetic studies focused on localizing and identifying the genes involved in adult-onset primary glaucoma, characterizing the gene products, and investigating the molecular mechanisms implicated in the pathophysiology of the disease.*
METHODS. *Several studies have aimed at understanding gene expression and protein processing and attempting to correlate the mutations identified in the involved genes, particularly the* TIGR/MYOC *gene, with the overall spectrum of the disease, ranging from juvenile glaucoma to typical late-onset primary open-angle glaucoma. Genetic research remains essential until highly specific and sensitive tests have been developed (plausible disease-causing sequence variations, polymorphisms).*
RESULTS. *The most effective method for detecting glaucoma clinically is the study of optic nerve and visual field damage, as well as intraocular pressure. In subjects at high risk, in members of families with a strong history of inherited glaucoma, and in families with a* MYOC*-positive test, the result may represent a marker to assess presymptomatic diagnosis and may be useful as a prognostic marker.*
CONCLUSIONS. OPTN *seems to have a role confined to the pathogenesis of normotensive glaucoma with a few exceptions. Presently, the introduction of the expensive and time-consuming* OPTN *gene test in the current diagnosis of familial glaucoma is not justified. (Eur J Ophthalmol 2004; 14: 220-5)*

KEY WORDS. *DNA analysis, Glaucoma genes, Myocilin, Optineurin, Primary open-angle glaucoma*

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It has long been observed that primary glaucoma tends to run in individuals belonging to the same families, but attempts to explain this fact in simple mendelian patterns of inheritance have so far failed. It was already noted that intraocular pressure (IOP) is in part genetically determined, and if it is true that elevated pressure is a cause of primary glaucoma, this could well represent one of the factors involved (1). In Armaly's theory (1), genetic determination represents the complex interactions of many pairs of alleles controlling the transmission of single parameters such as IOP, aqueous outflow facility, steroid hypertensive response, and optic cup; such a genetic determination and transmission seems to be absent in primary an-

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gle closure, secondary, and congenital glaucomas. Moreover, pigmentary glaucoma appears to be a separate entity, both etiologically and clinically (5). There are in literature several comprehensive surveys of the premolecular age studies of genetics (6-8).

Today, molecular genetics study has focused on localizing and identifying the genes involved in primary glaucoma, characterizing the gene products, and investigating the molecular mechanisms implicated in the pathophysiology of the disease. Much effort has been aimed at understanding the gene expression and protein processing and at attempting to correlate the mutations identified in the involved genes, particularly the TIGR/MYOC gene, with the overall spectrum of the disease, ranging from juvenile to typical late-onset primary open-angle glaucoma (POAG).

Juvenile open-angle glaucoma (JOAG) was the most promising starting point in this research. Sheffield et al (9) located a gene responsible for this condition at chromosome 1q21-q23: this locus, the first locus mapped by linkage analysis for open-angle glaucoma, was named GLC1A, according to the Human Genome Organization/Genome Database nomenclature (http://www.gene.ucl.ac.uk/hugo/). In 1997 (10), Stone et al reported the identification of a gene associated with GLC1A, called TIGR (trabecular meshwork-induced glucocorticoid response protein); the gene was found to code for a protein originally described by Polansky et al (11, 12), produced by trabecular meshwork and ciliary body cells in response to glucocorticoids, with a time scale similar to that observed in steroid-induced glaucoma. This protein, recently renamed myocilin (MYOC), is expressed in cells of the trabecular meshwork, in other ocular sites, and in many human tissues. In the trabecular meshwork the production of myocilin can be induced by the application of topical corticosteroids. Jacobson et al (13) have shown that normal myocilin is secreted into the medium and that there is a defect in the secretion of myocilin when myocilin mutants are transfected into cultured cells. These findings suggest that one possible function of myocilin is to regulate IOP.

Between 1996 and 1998, several other loci have been identified and associated with late onset glaucoma: on 2cen-q13 (GLC1B) (14), low to moderate IOP; on 3q21-24 (GCL1C) (15), high tension and visual field loss; on 8q23 (GCL1D) (16), moderate tension and visual field loss; finally, with normal tension glaucoma (NTG) on chromosome 10p15-14 (GCL1E) (17). However, it is debatable whether present knowledge allows such tight association. Moreover, in the field of glaucomas believed secondary, a locus has been identified and associated with glaucoma in pigment dispersion syndrome (PDS) on chromosome 7q35-36 (18). Ritch (19) reminds that one of the genes located at the telomeric end of chromosome 7q (20) is that for phenylthiourea tasters. This is interesting because the gene was long associated with POAG (the old PTC taste test, an associated nonocular marker (21). A second locus for the PDS has been suggested on 18q11-q21 (22), but not supported later. There are now two articles in which pigmentary patients have been found with myocilin mutations (23, 24). Consequently, it seems that pigmentary glaucoma and the so-called myocilin-related glaucoma are not always distinct entities. Until recently, pigmentary glaucoma has been considered nosologically either a congenital developmental glaucoma (25) or a congenital and environmental one (19), as well as a hypothetical variant of POAG (26) or as a secondary glaucoma (5). For the glaucoma in pseudo *exfoliatio lentis*, which also belongs to the secondary glaucomas, it has been suggested that there are at least two loci (27, 28), without further support. Approximately 2 to 4% of POAG patients carry a mutation of the MYOC gene in Colomb et al (29) and 2.6 to 4.3% in Fingert et al (30). Bonomi (31) estimates that the mutation is present in 8% of juvenile forms and in 5% of adult ones.

Widely variable phenotypes depending on the specific mutation in myocilin seem to exist. Besides the different loci that seem to lead to different characteristic forms of open-angle glaucoma, several mutations have been described in the GLC1A gene, and these can produce the disease with different clinical findings. Alward et al (24) enumerate at least 38 different mutations, with a minimum of 16 being disease-causing mutations. Few mutations seem to be more frequent and important: for instance, the nonsense mutation p.Q368X (Gln368Stop), which is related to a late onset phenotype and is of moderate severity. On the contrary, missense mutations, such as p.T377M (Thr377Met) and p.I477N (Ile477Asn), are related to early onset forms and are of much greater severity. We observed a new MYOC mutation in a two-generation family: a missense mutation p.C25R (Cys25Arg), which segregated with both POAG (the father) and JOAG

(the first son), but not with PDS (the second son), nor in the healthy daughter (32).

More recently, molecular geneticists have paid great attention to describing the prevalence and characterization of plausible disease causing sequence variations (DCVs) and to distinguishing pathogenic myocilin mutations from the apparently benign ones, or rather, from polymorphisms (24). Some criteria for judging such pathogenicity are as follows: physicochemical changes, statistical arguments (high prevalence or rarity of the mutation), and *in vitro* assays (33). In mutation-specific glaucoma phenotypes, it is clear that the clinical spectrum of the disease due to MYOC mutation can range from JOAG to typical late-onset POAG, as many articles suggest (34, 35) and, perhaps, to other types of glaucoma, with the exception of NTG (36). In Japan, Kitazawa (37) has observed myocilin mutations in only 0.5% of POAG patients and 2.37% of NTG patients, and concluded that POAG and NTG belong to an identical spectrum of diseases.

In the hope of finding useful indicators of effectiveness of treatments in the management of glaucoma patients (IOP control and visual field damage restraint), ophthalmologic interest has stimulated research to investigate hypothetical relationships between the severity of the disease and a genetic basis, such as the biallelic single nucleotide polymorphism in the promoter of the MYOC gene reported by Colomb et al (29), which leads these authors to comment that "*MYOC mt 1 typing could be a matter of public health*". In a much larger population, Alward et al (25) do not confirm the results of Colomb et al, and other authors stress that the relative commercial assay, the OcuGene Test, is too expensive (38).

Not all cases of JOAG or POAG have mutation in the MYOC gene, however, implicating other genes (note Quigley's conclusions concerning the leading role of the optic nerve in the disease) (39). In 2002, Rezaie et al (40) discovered another gene in the GLC1E interval on chromosome 10p. The gene was called OPTN and its protein product optineurin (for optic neuropathy inducing protein); the gene was previously identified as FIP-2 (41) and as NRP (42). Optineurin is expressed in ocular tissues, brain, and elsewhere. Optineurin interacts with proteins mutated in neurodegenerative diseases such as Huntington hereditary chorea; in addition, optineurin may be a component of TNFα pathway, which regulates programmed cell death. Mutations of the OPTN gene were found in 16.7% of 54 families with adult-onset POAG with at least one member with NTG; not enough is known about the detailed clinical aspects of the population studied. Recently, Walter et al (43) and Tang et al (44) denied that variations in the OPTN gene may cause or predispose individuals to adult-onset POAG. It seems that the population studied in Walter et al's work is different from the population studied by Rezaie et al. Tang et al form the hypothesis that there may be racial differences between Japanese and white subjects. Physiologic studies by Vittitow et al (45) point out that it is not yet clear how OPTN interacts with IOP mechanisms, other than to play the role of a general defense device against glaucomatous insult, in an unknown manner. We currently define glaucoma as a progressive optic neuropathy, shifting the emphasis from an IOP disease to a progressive degenerative disease of the optic nerve (according to Lichter (46), an intraocular pressure-sensitive neuropathy), where the most common known risk factor is the less or more elevated IOP. There are well-known non-IOP-dependent risk factors: perfusive, hemodynamic, and hemorheologic. The recently described OPTN gene must be considered as another risk factor.

Vincent et al (47) discovered a potential modifying effect on MYOC of the CYP1B1 gene (the gene mutated in about 85% of congenital glaucoma patients). Individuals with mutation in both genes developed POAG at a much younger age (mean 27 years) than did family members with mutation in MYOC alone (mean 51 years). This article affirms the digenic inheritance of early-onset glaucoma and emphasizes the genetic heterogeneity and complexity behind the pathogenesis of glaucoma.

Glaucoma has in common with other genetic diseases affecting retina, choroid, and vitreous the phenomenon by which a different mutation within the same gene can cause clinically distinct ocular diseases, defined as phenotypic diversity from allelic series, as, for instance, those of rhodopsin gene responsible for the autosomal dominant retinitis pigmentosa and the congenital stationary night blindness. In the phenomenon known as locus heterogeneity, instead, the same phenotype is produced by alterations of many different genes in different chromosomes, as typically in retinitis pigmentosa, as if the repertoire of responses of the eye to genetic anomalies could be limited.

Myocilin mutations were noted in only a few cases

of POAG. The low prevalence of myocilin-associated glaucoma precludes the value of mutation of the gene as a prognostic factor in POAG: it is premature to suggest the usefulness of the expensive genetic test. Nevertheless, in subjects at extremely high risk and in members of families with a strong history of inherited glaucoma or with a positive test, the result may represent a marker to assess presymptomatic diagnosis and may be useful as a prognostic marker. Genetic research and information remains essential until highly specific and sensitive tests are developed. Clinically, today the most effective method for detecting glaucoma is shill the study of optic nerve and visual field damage, as well as IOP.

At present, ophthalmologists need to improve their understanding of the molecular genetics of the regulation and production of aqueous humor and of factors that could modulate trabecular meshwork function (48). They also need an improved understanding of cell death pathways activated in retinal ganglion cells under various conditions, such as elevated and normal IOP. Molecular genetic studies need to provide more knowledge on the predictive power of mutations in genes – those already known and to be identified – of polymorphisms and on plausible disease-causing sequence variations in the myocilin gene and on other new disease-causing genes.

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