Pediatric cataract, myopic astigmatism, familial exudative vitreoretinopathy and primary open-angle glaucoma co-segregating in a family

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Purpose: To describe an Australian pedigree of European descent with a variable autosomal dominant phenotype of: pediatric cortical cataract (CC), asymmetric myopia with astigmatism, familial exudative vitreoretinopathy (FEVR), and primary open-angle glaucoma (POAG).

Methods: Probands with CC, FEVR, and POAG were enrolled in three independent genetic eye studies in Tasmania. Genealogy confirmed these individuals were closely related and subsequent examination revealed 11 other family members with some or all of the associated disorders.

Results: Twelve individuals had CC thought to be of childhood onset, with one child demonstrating progressive lenticular opacification. One individual had severe retinal detachment while five others had dragged retinal vessels. Seven individuals had POAG. Seven individuals had myopia in at least one eye ≤ -3 Diopters. DNA testing excluded mutations in myocilin, trabecular meshwork inducible glucocorticoid response (MYOC) and tetraspanin 12 (TSPAN12). Haplotype analysis excluded frizzled family receptor 4 (FZD4) and low density lipoprotein receptor-related protein 5 (LRP5), but only partly excluded EVR3. Multipoint linkage analysis revealed multiple chromosomal single-nucleotide polymorphisms (SNPs) of interest, but no statistically significant focal localization.

Conclusions: This unusual clustering of ophthalmic diseases suggests a possible single genetic cause for an apparently new cataract syndrome. This family’s clinical ocular features may reflect the interplay between retinal disease with lenticular changes and axial length in the development of myopia and glaucoma.

In this study, we describe the novel overlapping phenotype of congenital cataract (CC), familial exudative vitreoretinopathy (FEVR), myopia, and primary open-angle glaucoma (POAG) segregating in an apparently autosomal-dominant fashion.

In Australia, myopia affects approximately 15% of the population [1], POAG affects approximately 3% of the population [2], CC occurs in approximately 2.2 out of every 10,000 births [3], and FEVR affects an estimated 7 out of every 100,000 people (derived from comparing 13 indexed FEVR cases [4] to 420 CC cases [3]). If we were to consider these diseases as completely independent clinical entities, the highly unlikely probability of a patient having all four diseases simultaneously, or of the four diseases co-segregating, would be approximately 1 in 148 billion. This denominator is more than 20 times the total population of earth today.

Interestingly, to some extent these clinical entities can be associated with each other. Many investigators have reported the association of high myopia with cataract, glaucoma, and retinal detachment [5]. Other associations are less common:

• anterior polar cataracts, seen in aniridia, are often associated with glaucoma [6];
• rubella embryopathy is associated with both congenital glaucoma and CC [6];
• aphakic glaucoma is observed very frequently, and cataract can develop as a complication of POAG-filtering surgery [6];

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retinal detachment is a feature of Stickler syndrome and is associated often with cortical lens opacities [7];

retinal detachment from retinopathy of prematurity (ROP) is associated with myopia and cataract [8].

Retinal dystrophies are associated with myopia and posterior subcapsular cataracts [9].

Although researchers have identified genes associated with each of these disorders, the genetic mechanisms and their interactions still are not fully understood.

METHODS

We identified three closely-related index cases from three genetic-eye-disease studies: VI:7 from the Glaucoma Inheritance Study in Tasmania (GIST) [10], VIII:7 from the Cataract Inheritance Study in South Eastern Australia (CISSEA) [3], and VIII:8 from the Familial Retinal Detachment Study (FRDA) [4]. The GIST study had ethical approval from the Royal Hobart Hospital; the CISSEA and FRDA studies had ethical approval from the Royal Victorian Eye and Ear Hospital. In each case, the work was conducted in accordance with the tenets of the Declaration of Helsinki.

When we realized that the index cases were a grandmother and two of her grandchildren who were genetic first cousins, we decided to examine the entire pedigree in detail to characterize a potentially novel phenotype. Our ultimate aim was to identify the gene responsible for this apparently-autosomal-dominant disorder.

From the genealogy of the index cases [11] we identified the living members of five lineal generations, as well as surviving more-distant relatives. We invited these family members for a comprehensive ophthalmic examination [12], including:

Table 1. Microsatellite primers and conditions.

| Marker   | Primer names and sequences (5'-3')                  | Size (bp) | Annealing temperature | Amplification conditions |
|----------|----------------------------------------------------|-----------|-----------------------|--------------------------|
| D11S4187 | F TCTTGACCCCGGAAG R CGGTCGCTGTTGGA                 | 273-289   | 55 °C                 | Invitrogen Taq & buffer  |
| D11S896  | F AATCCTCTATGCTGTGGGA R AGTATCACCCTCACA             | 169-183   | 60 °C                 | Invitrogen Taq & buffer  |
| D11S1367 | F GCTGACATTATTCATGTGGG R ACAAGTTTATCTCCCTGGA       | 224-244   | 60 °C                 | Invitrogen Taq & buffer  |
| D11S2006 | F CTGTGGCGTGTGTTGTC R AAAGATATCAAGAAGATAAGG        | ~325      | 55 °C                 | Invitrogen Taq & buffer  |
| D11S4095 | F TCCCTGGATCTCTGAATC R CTGTGCTGGTCCAG              | 173-205   | 55 °C                 | Invitrogen Taq & buffer  |
| D11S937  | F CTAATAAACAATCCCTACCTCC R TATGTCAGTCAGGAAGCCAAATG| 230-264   | 60 °C                 | Invitrogen Taq & buffer  |
| D11S929  | F AGGCCTTCAAGCTACG R CCAGTGCTGGAGAACC              | 218-240   | 60 °C                 | Invitrogen Taq & buffer  |
| D11S4115 | F TGGGATGTATAATAGAGAAGATAC TGTGCATCAGAGTAAC        | 185-199   | 50 °C                 | Invitrogen Taq & buffer  |
| D11S4154 | F ATCCCTGTCTTTCTAGAGCAC R GTGGCTCTCAACCTCAGT        | 146-158   | 65 °C                 | Invitrogen Taq & buffer  |
| D11S4203 | F GAATAGCCACTGACTCAAG R CAGGAATGCTGGAATAGAGAA      | 218-278   | 60 °C                 | Invitrogen Taq & buffer  |
| D11S4083 | F TTAAACCAAGGAAGG CAGAC R CATGTGTCACCAAGGGCAG      | 178-206   | 55 °C                 | Invitrogen Taq & buffer  |
| D11S4102 | F CACCACTGGGTACTGGCATT CAG R GCTAAATCTCGGAAAAGCTCTG| 142-174   | 60 °C                 | Invitrogen Taq & buffer  |

Table 2. TSPAN12 primers and PCR conditions.

| Exon | Primer names and sequences (5'-3')                  | Size (bp) | Annealing temperature | Amplification conditions |
|------|----------------------------------------------------|-----------|-----------------------|--------------------------|
| 2    | TSPAN12-ex2-F ATGTCGCCGTGCCTCTCTCC                 | 382       | 60 °C                 | Invitrogen Taq & buffer  |
|      | TSPAN12-ex2-R CCAGGGTTGTTTCTCTTCTTTGT              |           |                       |                          |
| 3    | TSPAN12-ex3-F TGTTATGCGGCAGAGTATATAGTAAC           | 291       | 60 °C                 | Invitrogen Taq & buffer  |
|      | TSPAN12-ex3-R CCAGGGTTGTTTCTCTCCTG                 |           |                       |                          |
| 4    | TSPAN12-ex4-F TGAGGCATCATGTTGGAAAGAAGA             | 346       | 60 °C                 | Invitrogen Taq & buffer  |
|      | TSPAN12-ex4-R GGCTATCTGCCTGGCCATAGGTGTC           |           |                       |                          |
| 5    | TSPAN12-ex5-F GGTGCCCTTCCTTGGGAGAAGA              | 947       | 60 °C                 | Invitrogen Taq & buffer  |
|      | TSPAN12-ex5-R TCGAAATGTTCTACGCAGCAG               |           |                       |                          |
| 6    | TSPAN12-ex6-F GTACAAAATCCTCCTTATTTATCATCA          | 529       | 60 °C                 | Hot shot master mix      |
|      | TSPAN12-ex6-R GGAAGAAAAGGGAGAGAAAGG               |           |                       |                          |
| 7    | TSPAN12-ex7-F TATGGCAGACATATAGCCTGTGG             | 376       | 60 °C                 | Hot shot master mix      |
|      | TSPAN12-ex7-R TTTTTAAGGGCTTTTTACATTGTAGAC         |           |                       |                          |
| 8    | TSPAN12-ex8-F GTCTTTCCCTGAAGACACCCTG              | 605       | 60 °C                 | Hot shot master mix      |
|      | TSPAN12-ex8-R CCATCTCCATTTTTAAGCATAG              |           |                       |                          |
•a LogMAR visual acuity test,
•the Goldmann applanation intraocular pressure (IOP) measurement,
•refraction using a HARK-598 autorefractor (Carl Zeiss Meditec, Miami, FL),
•axial length measurement using an Ocuscan® (Alcon, Inc., Ft Worth, TX),
•corneal pachymetry using an IOPac (Heidelberg Instruments, Heidelberg, Germany),
•lens photographs,
•stereoscopic optic disc photography using a Nidek 3Dx camera (Nidek, Gamagori, Japan), and
•examination of the peripheral retina.

All participants provided venous blood or saliva specimens for DNA extraction and genetic analysis.

Genotyping was performed using fluorescently-tagged microsatellite markers as described previously [13]. Briefly, standard PCR reactions were carried out in a 25 μl volume containing 50 ng of genomic DNA using Invitrogen Taq DNA polymerase and buffers (Invitrogen). Microsatellite markers (including primer details; Table 1) surrounding EVR1 (D11S4187, D11S896, and D11S1367), EVR4 (D11S2006, D11S4095, and D11S937) and EVR3 (D11S929, D11S4115, D11S4154, D11S4203, D11S4083, and D11S4102) were selected from the genome browser. Following amplification, PCR products were resolved using an ABI 3730 DNA sequencer and analyzed using GeneMapper® software from the same manufacturer (Applied Biosystems, Carlsbad, CA).

The coding sequence and surrounding exons of myocilin, trabecular meshwork inducible glucocorticoid response (MYOC) and tetraspanin 12 (TSPAN12; primers and conditions are listed in Table 2) were screened using standard direct sequencing protocols as described previously (see above) [14,15].

For the genotyping platform, we used Linkage Panel IVb of 6008 genome-wide single-nucleotide polymorphisms (SNPs; Illumina, San Diego, CA), and ran the analysis at the Center for Inherited Disease Research (CIDR) of Johns Hopkins University (Baltimore, MD). The results for the pedigree were analyzed with Fastlink using a 2-point analysis (under a dominant model); multipoint results (both parametric and non-parametric) were analyzed using MERLIN. Merlin (Multipoint Engine for Rapid Likelihood Inference) is a software package that uses sparse inheritance trees for pedigree analysis [16].

RESULTS

Genealogical information was available for nine generations of the participants’ family; the individuals examined for this study came from the five most recent generations.

•Figure 1 shows the relevant portions of the full pedigree. A consanguineous loop enriched the pedigree with similar genes (RELPAIR [17] analysis suggested a grandparent–grandchild relationship when they were actually great-grandparent and great-grandchild).

•Table 3 displays the participants’ ophthalmic phenotypes with autorefraction sphere and cylinder, Keratometry readings, and axial length.

•Figure 2 and Figure 3A-N show photos of the optic disc, retina, and lens.

•Figure 4A-E show visual field defects.

Excluding the married-in spouses, we examined eight female and six male family members aged 3–86 years who apparently were affected.

•Visual acuity ranged from 6/5 to perception of light.
•Spherical-equivalent refractive error in Diopters (D) ranged from +0.25 D to −11.0 D, with five individuals having myopia in at least one eye of <-3D.
•Astigmatism varied from 0 to −7.25 D with the rule or −5 D against the rule.
•Axial length varied from 23.75 mm to 26.77 mm.
•Keratometry readings in eyes that had not been operated on ranged from 40.0 D to 48.62 D, with the largest corneal astigmatism measuring only 3.12 D.
•Maximum recorded IOP ranged from 13 mmHg to 36 mmHg.
•Central corneal thickness ranged from 510 μm to 590 μm.
•One male (VIII:6) was found to have a distance exotropia of 25 D.

One female individual (VIII:8) had severe spontaneous retinal detachment consistent with FEVR, while five individuals (3 male and 2 female) had dragged retinal vessels (V:4,V:1:7, VII:5, VII:7, VIII:3, VIII:6, VIII:7, IX:1). The youngest age of documented cataract was 3 years of age (IX:1).

One member (VIII:7) had photographic evidence of cataract progression (Figure 3J,K). In addition, iris atrophy was noted at the 3 and 9 o’clock positions. This atrophy possibly became more notable with age (Figure 3K).

One female individual (VIII:8) had severe spontaneous retinal detachment consistent with FEVR, while five individuals (3 male and 2 female) had dragged retinal vessels (V:4,VI:1:7, VII:5, VII:7, VIII:7, VIII:8).
Seven individuals (5 female and 2 male) had been diagnosed with POAG (V:2, V:4, VI:7, VI:12, VI:13, VII:5, VII:7).

Cataract extraction was performed on VII:7 after the cortical wedge progressed to complete lenticular opacification in the left eye and vision declined from 6/18 to 6/60. Post-operatively, this member’s best-corrected visual acuity improved to 6/6. Refraction in the left eye changed from −6.25/-1.5×145 to +0.00/-0.50 X 98 following cataract surgery. The brother of this individual (VII:5) had similar surgery for cataract and astigmatism, but his visual acuity did not improve from 6/60.

Systemic associations: None of the family members had dysmorphism or an unusual stature consistent with the facial or body habitus features of Stickler syndrome. One member, who had not worn ear protection in his industrial employment, had noise-related hearing loss (VII:7) and one (V:4) had age-related hearing loss. Only one member (V:4) was found to have a single café-au-lait spot.

One participant (VII:7) had previously been diagnosed with pulmonary alveolar proteinosis (PAP) and treated with repeated pulmonary lavage. PAP is a rare disorder related to the receptor pathway of the granulocyte macrophage–colony stimulating factor (GM-CSF); it was diagnosed after recurrent bouts of pneumonia in adult life. No other family member has
| ID   | Sex | Age at initial examination (years) | Refractive error (D) | Keratometry (D) | Axial Length (mm) | Cataract | Glaucoma | Dragged disc or retinal detachment |
|------|-----|----------------------------------|----------------------|-----------------|-------------------|-----------|----------|-----------------------------------|
|      |     |                                  | Right               | Left            | Right             | Left      |          |                                   |
| V:2  | F   | 81                               | -0.75/-2.00x70      | -0.75/-1.5x65   | 45.3/44.3         | 43.8/42.9 | Yes      | Yes                 | None         |
| V:4  | F   | 83                               | -2/-3.25x45         | -3.25/-1.0x95   | 49.2/53.9*        | 48.6/46.2 | Yes      | Yes                 | Dragged disc |
| V:7  | F   | 65                               | -0.25/-3.00x180     | -0.25/-3.5x75   | 45.3/42.25*       | 45.8/43.0 | Yes      | Yes                 | Dragged disc |
| V:12 | F   | 76                               | NR                  | NR              | NR                | NR        | Yes      | Yes                 | None         |
| V:13 | F   | 86                               | NR                  | NR              | NR                | NR        | No       | Yes                 | None         |
| VII:1| M   | 45                               | 0/-0.25x180         | 0/-0.25x180     | 40.043.1          | 43.4/44.3 | Yes      | No                  | None         |
| VII:3| F   | 43                               | 0/-1.5x180          | 0/-0.25x160     | NR                | NR        | Yes      | No                  | None         |
| VII:5| M   | 39                               | -3.25/-4.0x180      | -0.5/-1.0x175   | 43.5/45.1         | 43.5/43.6 | Yes      | Yes                 | Dragged disc |
| VII:6| F   | 33                               | -0.76               | -0.5/-0.75x170  | NR                | NR        | No       | No                  | None         |
| VII:7| M   | 36                               | -2.25/-0.5x155      | -0.5/-0.5x145   | 42.1/42.3         | 43.5/43.1 | Yes      | Yes                 | Dragged disc |
| VII:8| F   | 38                               | NR                  | NR              | NR                | NR        | No       | No                  | None         |
| VIII:1| F | 25                              | -0.25/-0.5x88       | 0/-0.25x302     | NR                | NR        | Yes      | No                  | None         |
| VIII:5| M | 16                              | -1.75/-1.25x50      | -0.5/-0.5x100   | 40.043.1          | 43.4/44.3 | Yes      | No                  | None         |
| VIII:6| M | 12                              | -2.0/-0.5x90        | -0.5/-0.25x95   | NR                | NR        | Yes      | No                  | None         |
| VIII:7| M | 8                               | -5.75/-0.5x55       | -9.25/-3.5x95   | 41.041.8          | 40.0/41.0 | Yes      | No                  | Dragged disc |
| VIII:8| F | 17                              | +0.5/-0.5x155       | -9.25/-3.5x95   | NR                | NR        | Yes      | No                  | Dragged disc |
| VIII:9| F | 15                              | -0.25/-0.5x135      | 0/-0.25x55      | NR                | NR        | No       | No                  | None         |
| IX:1 | M   | 3                               | 1.5                 | 0/-0.5x121      | 45.3/44.3         | 43.8/42.9 | No       | Yes                 | None         |

Abbreviations: F, female; M, male; D, diopters; NR, not recorded; ND, not determinable; OU, both eyes. *measured following cataract surgery
experienced similar medical problems; no individual reported any renal problems.

*MYOC* screening of the index case revealed no mutation [14]. Haplotype analysis of a central portion of the pedigree excluded the *EVR1* frizzled family receptor 4 (*FZD4*) and *EVR4* low density lipoprotein receptor-related protein 5 (*LRP5*) *FEVR* genes (Figure 4). Unfortunately, the *EVR3* locus could be only partially excluded due to uninformative markers. Given that this gene had not been identified, we cannot exclude this locus fully. Direct screening of VIII:8 excluded the recently-identified FEVR gene *TSPAN12*.

The family was included in the International High Myopia Consortium linkage analysis [16]; however, the family was dropped from the multipoint analyses for
chromosomes 3, 4, 6, 7, 8, 11, and 12 due to the pedigree's complexity. Table 4 displays the two-point linkage results for this family showing the highest scoring logarithm of odds (LOD) scores above 1.5. There were multiple chromosomal SNPs of interest, but no statistically significant focal localization.

DISCUSSION

This Australian pedigree has a unique constellation of ophthalmic features that do not appear to have been described previously. Although we were unable to identify a similar family reported in the literature, the subtle and relatively common clinical features could be overlooked.

Many investigators have reported the association of high myopia with ocular morbidity of early-onset cataract, glaucoma and retinal detachment [5]. Pedigrees with myopia are common, but pedigrees with so many members affected with these early ocular issues along with myopic development are extremely rare; we were not able to identify any in the published literature.

Although we cannot discount that the associated ocular features may be secondary in origin, this family raises the possibility that the same gene may be responsible for all forms of the pathology observed in the pedigree.

Retinal detachment is an uncommon disorder in young people and is most commonly identified in patients with FEVR. X-linked FEVR and Norrie disease arose from mutations in Norrin (excluded by male-to-male transmission, in this pedigree). Dominant FEVR is due to mutations in FZD4 and LRP5, and has been linked to the EVR3 locus [18]. We excluded these loci through linkage analysis. The recently-described gene TSPAN12 (EVR5) was excluded by sequence analysis. Nonetheless, despite a well characterized FEVR mutation, there still can be considerable variation in the expressivity of the phenotype and incomplete penetrance [15,18,19] (Personal communication; T.L. Edwards, Centre for Eye Research Australia, Melbourne, Australia [article in press]).

Since the cataract is the most “easily characterized” phenotype in this family’s pedigree, we compared it with other cataract phenotypes described in the literature. Although CC has been linked to or associated with many cataract loci and many chromosomal deletions, the causative mutation has not
Table 4. Summary of the Johns Hopkins Center for Inherited Disease Research (CIDR) results for the family.

| Chromosome | Marker | Position (cM) | 2PT-parametric (Fastlink) | MPT-non-parametric | MPT-parametric |
|------------|--------|---------------|---------------------------|--------------------|----------------|
| 1          | rs1981193 | 121.82        | 1.863                     | NS                 | NS             |
| 1          | rs1806753 | 160.34        | 1.079                     | NS                 | NS             |
| 2          | rs2053372 | 47.98         | 1.592                     | NS                 | NS             |
| 2          | rs2008535 | 54.9          | 1.128                     | NS                 | NS             |
| 2          | rs764464  | 65.31         | 1.328                     | NS                 | NS             |
| 2          | rs1022298 | 117.27        | 1.162                     | NS                 | NS             |
| 2          | rs264963  | 117.39        | 1.162                     | NS                 | NS             |
| 3          | rs2076993 | 46.5          | 1.166                     | NS                 | NS             |
| 3          | rs1348979 | 49.44         | 1.166                     | NS                 | NS             |
| 3          | rs1127732 | 59.51         | 1.097                     | NS                 | NS             |
| 3          | rs713144  | 60.4          | 1.477                     | NS                 | NS             |
| 3          | rs1382554 | 60.41         | 1.097                     | NS                 | NS             |
| 3          | rs1405793 | 64.61         | 1.159                     | NS                 | NS             |
| 3          | rs1495704 | 65.68         | 1.159                     | NS                 | NS             |
| 3          | rs1995137 | 66.29         | 1.159                     | NS                 | NS             |
| 3          | rs1351631 | 67.73         | 1.522                     | NS                 | NS             |
| 3          | rs737516  | 67.73         | 1.522                     | NS                 | NS             |
| 3          | rs1013758 | 67.81         | 1.522                     | NS                 | NS             |
| 3          | rs844438  | 78.91         | 1.123                     | NS                 | NS             |
| 3          | rs1447971 | 82.11         | 1.842                     | NS                 | NS             |
| 3          | rs935734  | 92.98         | 1.586                     | NS                 | NS             |
| 3          | rs1019374 | 95            | 1.069                     | NS                 | NS             |
| 3          | rs1388276 | 99.96         | 1.116                     | NS                 | NS             |
| 4          | rs751266  | 67.19         | 1.054                     | NS                 | NS             |
| 4          | rs896656  | 93.96         | 1.326                     | NS                 | NS             |
| 4          | rs2203837 | 23.58         | 1.615                     | NS                 | NS             |
| 8          | rs334206  | 32.33         | 1.241                     | NS                 | NS             |
| 8          | rs241202  | 48.58         | 1.849                     | NS                 | NS             |
| 8          | rs4107736 | 50.87         | 1.248                     | NS                 | NS             |
| 8          | rs1481747 | 53.13         | 1.103                     | NS                 | NS             |
| 8          | rs1955185 | 61.16         | 1.05                      | NS                 | NS             |
| 8          | rs716583  | 65.56         | 1.116                     | NS                 | NS             |
| 8          | rs344278  | 74.88         | 1.582                     | NS                 | NS             |
| 8          | rs1460239 | 112.26        | 1.618                     | NS                 | NS             |
| 8          | rs1433396 | 122.14        | 1.119                     | NS                 | NS             |
| 8          | rs766811  | 138.68        | 1.16                      | NS                 | NS             |
| 9          | rs1532310 | 0.124137      | 1.522                     | NS                 | NS             |
| 9          | rs1532309 | 0.124434      | 1.522                     | NS                 | NS             |
| 9          | rs1143025 | 30.9          | 1.176                     | NS                 | NS             |
| 9          | rs1029015 | 35.12         | 1.767                     | NS                 | NS             |
| 9          | rs716933  | 60.37         | 1.089                     | NS                 | NS             |
| 9          | rs987187  | 60.4          | 1.128                     | NS                 | NS             |
| 9          | rs1333342 | 69.96         | 1.477                     | NS                 | NS             |
| 10         | rs1346300 | 75.86         | 1.522                     | NS                 | NS             |
| 11         | rs676943  | 125.79        | 1.015                     | NS                 | NS             |
| 12         | rs871880  | 58.31         | 1.123                     | NS                 | NS             |
| 12         | rs7134835 | 161.7         | 1.2                       | NS                 | NS             |
| 12         | rs1278602 | 171.56        | 1.089                     | NS                 | NS             |
| 12         | rs1278601 | 171.57        | 1.089                     | NS                 | NS             |
| 12         | rs937538  | 171.78        | 1.094                     | NS                 | NS             |
been identified for the majority of CC and pediatric cataract cases [6].

The peripheral cortical lamella wedge seen in this family is similar to that observed in Stickler syndrome [7] and also with neurofibromatosis Type 2 (NF2) [20]. Interestingly, one case describes NF2 associated with posterior subcapsular cataract and dragged disc [21]. In a series of 15 other NF2 patients, 12 patients had an epiretinal membrane in the macular or paramacular area and 11 patients had central posterior cortical, subcapsular, or peripheral cortical lens opacities [22]. NF2 arises from mutations in the Merlin gene on chromosome 22q12.2 [23].

The one case of PAP [24] prompted an investigation of possible genes involved in the GM-CSF pathway using the Online Mendelian Inheritance in Man® (OMIM) database at Johns Hopkins University. Of three loci associated with PAP, one gene located at chromosome 22q12.2-q13.1, Granulocyte-macrophage Colony-stimulating factor receptor, beta (CSF2RB) is adjacent to Merlin. Notably, on reviewing myopia loci, the myopia linkage found by Stambolian and colleagues [25] for marker D22S685 lies in chromosome region 22q12. This region has also been replicated in the Beaver Dam Eye study [26].

The refractive error recorded in this pedigree is atypical; most hereditary myopia is symmetric and usually is not associated with high astigmatism. To date there has been little investigation of the genetics of astigmatism, though genetic factors are likely to play a role [27]. It would appear that the myopia in this family originates in increased axial length rather than in the more usual primary lenticular fault. The degree of astigmatism in severely affected members, however, appeared to be both lenticular and corneal, suggesting a common mechanism of growth or compensation. The causative interaction of the cataract and the increased

| Chromosome | Marker   | Position (cM) | 2PT-parametric (Fastlink) | MPT-non-parametric | MPT-parametric |
|------------|----------|---------------|---------------------------|--------------------|----------------|
| 13         | rs2985981| 49.25         | 1.004                     | NS                 | NS             |
| 13         | rs2031836| 115.73        | 1.003                     | NS                 | NS             |
| 15         | rs1435735| 46.31         | 1.799                     | NS                 | NS             |
| 15         | rs890153 | 46.31         | 1.554                     | NS                 | NS             |
| 15         | rs725463 | 60.22         | 1.043                     | NS                 | NS             |
| 15         | rs1445020| 71.05         | 1.049                     | NS                 | NS             |
| 16         | rs1019141| 19.98         | 1.49                      | NS                 | NS             |
| 16         | rs889593 | 122.83        | 0.018                     | 0.701998           | 1.0217         |
| 16         | rs299956 | 123.93        | 0.734                     | 0.943619           | 1.5971         |
| 16         | rs2076962| 125.29        | −0.036                    | 1.127055           | 1.8771         |
| 16         | rs3794668| 126.97        | −0.011                    | 1.126755           | 1.8763         |
| 16         | rs1056707| 128.94        | 0.057                     | 1.12803            | 1.8782         |
| 16         | rs750740 | 129.03        | 0.399                     | 1.128125           | 1.8783         |
| 16         | rs463701 | 130.14        | −0.067                    | 1.129806           | 1.8804         |
| 16         | rs452176 | 130.21        | 0.01                      | 1.129825           | 1.8804         |
| 16         | rs1006547| 130.48        | 0.018                     | 1.129924           | 1.8805         |
| 16         | rs1800330| 130.5         | 0.891                     | NS                 | NS             |
| 16         | rs870856 | 130.83        | 1.781                     | 1.126244           | 1.8762         |
| 16         | rs8577   | 130.86        | 0.549                     | 1.125715           | 1.8755         |
| 17         | rs721429 | 95.95         | 1.199                     | NS                 | NS             |
| 18         | rs1972602| 45.77         | 1.123                     | NS                 | NS             |
| 18         | rs1548755| 51.57         | 1.252                     | NS                 | NS             |
| 18         | rs1131709| 56.82         | 1.339                     | NS                 | NS             |
| 18         | rs650680 | 58.25         | 1.767                     | NS                 | NS             |
| 18         | rs931078 | 84.57         | 1.11                      | NS                 | NS             |
| 20         | rs1535382| 14.16         | 1.046                     | NS                 | NS             |
| 21         | rs1041756| 33.98         | 1.07                      | NS                 | NS             |
| 21         | rs2839576| 62.26         | 1.324                     | NS                 | NS             |

2-point analyses with Fastlink under a dominant model; multipoint results, both parametric and non-parametric, using the multipoint engine for rapid likelihood inference (MERLIN). Results in italics highlight suggestive loci, while the results in bold were found to be suggestive under all models tested. Abbreviations: Chr, chromosome; cM, centimorgan; 2PT, two point; MPT, multi-point; NS, not significant.
myopia remains to be elucidated, but may involve visual form deprivation [28].

We hope that characterization of this unusual phenotypic constellation will identify other families with similar characteristics. Further characterization of the genes involved in this family using methods such as next-generation sequencing should help shed light on the genetics of the four clinical entities—POAG, CC, FEVR, and myopia—as well as their interactions. In time, this further work also may help clarify the molecular pathways of developing myopia involving retinal signaling, lens growth and axial length.

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