Juvenile bilateral lens dislocation and glaucoma associated with a novel mutation in the fibrillin 1 gene

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Purpose: To describe the clinical, ocular, and genetic findings in multiple members of a family with early-onset and bilateral lens dislocation, clinical corneal guttae, and glaucoma.

Methods: All family members underwent complete physical and ophthalmic examinations. After informed consent was given, DNA was obtained from eleven family members, eight of whom were affected. Three polymorphic markers near the fibrillin 1 (FBN1) locus were genotyped and the results analyzed using the VITESSE program. Amplification of the 65 exons and flanking intronic sequences of FBN1 was performed using polymerase chain reaction (PCR), followed by conformation sensitive gel electrophoresis (CSGE). Then, all fragments with mobility variations were sequenced.

Results: Pedigree analysis revealed a three generation family with eight of eleven individuals affected by early onset lens dislocation, high myopia, typical facies, frontal bossing, flexion contractures, proximal interphalangeal (PIP) joint thickening, clinical corneal guttae, and glaucoma. Genetic linkage analysis using polymorphic markers near FBN1 demonstrated an LOD score of 1.78 (maximum possible LOD score 1.78). Conformation sequence gel electrophoresis analysis suggested a sequence variation in exon 3. Sequencing revealed a C965G substitution, resulting in an S322C coding change. This sequence variant segregated with affection status and was not identified in 154 control chromosomes.

Conclusions: This syndrome is consistent with a novel mutation in the FBN1 gene. FBN1 mutations have been previously described as causative for Marfan syndrome. The early-onset of complete lens dislocation, progressive corneal guttae, and glaucoma is unusual for Marfan syndrome. This study expands the Marfan phenotype and demonstrates a possible link between guttae, glaucoma, and fibrillin 1 disorders.

Marfan syndrome (OMIM 154700) is associated with a tall stature, long limbs, pectus deformity, flexible joints, aortic root dilatation, and dissecting aneurysms. Fifty to eighty percent of patients with Marfan may have lens subluxation; however, lens dislocation is relatively rare. Inheritance is usually autosomal dominant with fifteen percent occurring as sporadic cases without a family history of Marfan syndrome [1,2].

Spontaneous lens dislocation has been associated with several inherited disorders all of which presumably involve defects in zonular structure or zonular attachment to the lens capsule. Homocystinuria is an autosomal recessive condition associated with deficient collagen I cross linking due to decreased cystathione β-synthase activity [3,4]. Sulfite oxidase deficiency is an autosomal recessive condition involving a disorder of sulfur metabolism due to mutations in the sulfite oxidase gene [5,6]. Familial ectopia lentis and ectopia lentis et pupillae have been reported in both autosomal dominant and recessive forms [7-9]. Weill-Marchesani syndrome has autosomal dominant and recessive forms. The dominant form localizes to 15q21.1, producing a defect in fibrillin-1 or microfibril-associated proteins. The recessive form localizes to 19p13.3-p13.2 and has been reported to be associated with mutations in the ADAMTS10 gene [10]. Patients typically have microspherophakia with brachydactyly, stiff joints, and short stature [11].

Here we present a family with an autosomal dominant form of juvenile lens dislocation not associated with the usual phenotypical findings of the other syndromes listed above. We describe three generations of a family affected with lens dislocation, clinical corneal guttae, glaucoma, flexion contractures of the elbow, proximal interphalangeal (PIP) joint prominence, high arched palate, and pectus deformity of the chest.

METHODS

Duke University IRB approval and informed consent were obtained on all family members. All eleven individuals underwent a complete ophthalmic and physical examination and provided blood samples for DNA extraction. Family members were genotyped for polymorphic markers D15S1232, D15S659 and D15S648 using the FASST method [12]. Simulations of maximum possible LOD score were performed using the SIMLINK program [13] and linkage analysis was performed using VITESSE [14]. The fibrillin-1 gene was screened for mutations; all 65 exons were amplified from genomic DNA using PCR and analyzed using conformation sensitive gel electrophoresis (CSGE). Fragments with mobility variations were sequenced using an ABI PRISM 3100 Sequencer (Beverly, MA).
RESULTS

Clinical findings: The proband was a product of a full term, uncomplicated pregnancy of nonconsanguineous parents. He is a 36-year-old white male with a history of normal vision in early childhood but developed visual difficulties around six years old. He received aphakic spectacle correction by age eight years and by age 26, his intraocular pressures (IOP) were found to be elevated to around 35 mmHg. Medical treatment was initiated at 30 years old and pressures were maintained in the low 20 mmHg range. He had been treated for glaucoma for approximately ten years and re-presented with a complaint of intermittent ocular pain. Examination revealed pressures of 39 mmHg OD and 52 mmHg OS without observable pupillary block. His medical regimen was advanced and pressures again maintained in the low 20 mmHg range. He was then referred to our institution for further management.

On presentation, best-corrected visual acuity was 20/50+2 OD and 20/40+1 OS. Biomicroscopic examination revealed moderate corneal guttae without visible corneal edema and the anterior chambers were deep without any evidence of active inflammation. No cell or flare was present and both irides demonstrated frank iridodonesis. Gonioscopy revealed approximately four clock hours of broad peripheral anterior synchiae in the right eye and three clock hours in the left. The remaining angle structures in each eye appeared normal with visibility to the level of the scleral spur. Intraocular pressures were 24 mmHg in each eye. Funduscopic examination demonstrated healthy and pink 0.45 cups OU with a normal-looking macula and vessels. Completely dislocated lenses with intact lens capsules were present in the vitreous cavity of each eye. Ultrasonography confirmed the above lens findings (Figure 1). Systemic examination demonstrated a short forehead with midface hypoplasia and a prominent jaw. A pectus deformity of the chest was present along with PIP joint prominence and dolichostenomelia with joint stiffness (Figure 2). Table 1 summarizes the findings in all affected individuals.

The proband’s mother (Case I:2) was the product of a full term, uncomplicated pregnancy of nonconsanguineous parents. She was noted to have poor eyesight since early childhood. She recalls a history of frequent “pressure pain” OU with loss of all vision OD by 10 years of age. At age 26, she frequently perceived “yellow halos” with pain and progressively deteriorating vision OS. Her first ophthalmic examination was around 30 years of age at which time she had no light perception (NLP) vision OU documented. On presentation, she was 75 years old with NLP vision OU with nonreactive pupils. She exhibited a 30 prism diopter exotropia but extraocular motility was full. Slit lamp examination revealed normal appearing conjunctivae OU. Corneal exam showed central corneal stromal opacities and corneal edema. Early band keratopathy was present OU. Gonioscopy, optic nerve head, and fundus examinations were unable to be performed due to poor visibility of these structures. Ultrasound examination revealed findings consistent with lens remnants in the vitreous cavity. Systemic examination revealed similar findings to the proband. Cardiac catheterization revealed no evidence of aortic root dilation.

Figure 1. B scan ultrasound of the proband’s (II:1) right eye demonstrating complete lens dislocation into the vitreous cavity. All eight affected individuals in this pedigree similarly demonstrated complete lens dislocation.
The findings of the remaining family members are summarized in Table 1 and Table 2. The oldest child of the proband underwent an echocardiogram that revealed a dilated aortic root. He was diagnosed with Marfan syndrome by the Ghent criteria [15]. For this reason, we evaluated linkage in this family to the causative gene for Marfan syndrome, FBN1.

Polymorphic markers D14S1232, D15S659 and D14S268 surrounding the fibrillin 1 gene were genotyped in this pedigree. Simulation studies (SIMLINK) of an affected’s only analysis using equal allele frequencies and assuming equal recombination rates in males and females demonstrated a maximum possible two-point LOD score of 1.78 with the mean score of 0.81. An affected’s only analysis (VITESSE) with a fully penetrant, autosomal dominant model revealed a two-point LOD score of 1.78 for markers D15S659 and D15S648. Figure 3 demonstrates a haplotype that segregates with disease in this pedigree, supporting a role of variants in the FBN1 gene in the disease.

CSGE analysis suggested a sequence variation in exon 3 of the FBN1 gene. Sequence analysis revealed a heterozygous missense mutation (C965G) resulting in a Ser322Cys amino acid substitution. This sequence variant was found to segregate completely with the phenotype, and was not detected in 154 control chromosomes.

**DISCUSSION**

We describe three generations of a family with a syndrome consisting of juvenile bilateral lens dislocation, glaucoma, and the clinical appearance of corneal guttae. These findings segregate with a heterozygous missense mutation in exon 3 of FBN1. The first affected member appears to be the mother of the proband since she has no recollection of any older relatives with any similar ocular or systemic stigmata. The disorder has complete co-segregation of genotype and phenotype. Fibrillin 1 genetic changes have been associated with Marfan syndrome [16], Marfan related syndromes [17,18], and autosomal dominant Weill-Marchesani syndrome [11]. The typical features of Weill-Marchesani syndrome are absent in this family and this family meets the Ghent criteria for Marfan syndrome given the findings in one of the proband’s children.
Therefore, this mutation appears to produce a variant of Marfan syndrome with the striking feature of a juvenile onset lens dislocation.

Pedigree analysis demonstrated autosomal dominant inheritance with high penetrance (Figure 3). The S322C mutation leads to the gain of a cysteine residue in a calcium-binding epidermal growth factor-like (EGF-like) domain of fibrillin 1. Although many more Marfan mutations are associated with the loss of a cysteine residue, there are several prior reports of a disease-associated cysteine gain as seen in this family [16,19,20]. Cysteine residues are highly conserved in fibrillin 1 and are intimately involved in the tertiary structure of the fibrillin 1 protein due to the disulfide bonds they form. They are the major structural element of the 10-12 nm diameter connective tissue microfibrils which provide structural support for connective tissues throughout the body [21]. Therefore fibrillin 1 mutations presumably lead to a change in the function or stability of fibrillin-1 and phenotypically display connective tissue derangement. The exact pathogenesis of the mutation described in this family is unknown; however, both haploinsufficiency and dominant-negative models have been suggested for heterozygous missense fibrillin 1 mutations. Animal model studies have suggested that some missense mutations can lead to retention of the mutant fibrillin 1 in the endoplasmic reticulum and produce a haploinsufficiency effect [21,22]. Other missense mutations can lead to normal extracellular transport of the protein and presumably lead to deficient cross-linking and hence have a dominant-negative effect [21]. Moreover, another possible effect of this mutation could involve increased local clearance of mutant fibrillin 1. The bound calcium in the calcium-binding EGF-like domains of fibrillin 1 provide increased rigidity to the tertiary structure of the protein and protect it from proteases [23,24]. Therefore, derangement of such a calcium binding domain could lead to an effective extracellular haploinsufficiency.

A leading feature of this family is the unusual presentation of early lens dislocation. Involvement of the fibrillin-1 gene suggests that zonular instability is a primary factor leading to lens dislocation. Zonules suspend the lens from the ciliary body and are composed of microfibrils whose major components are fibrillin-1 and fibrillin-2 proteins [25-28]. Several identified mutations, R62C, S115C, R122C, R240C, R545C, R627C, and R1530C result in cysteine substitutions with associated ocular manifestations [20]. However, none have reported lens dislocation as a primary ocular phenotype. In general, complete lens dislocation is relatively rare in Marfan syndrome and to our knowledge this is the first reported family with bilateral juvenile onset lens dislocation.

Glaucoma is highly prevalent in this family with 5 of 8 confirmed affected individuals. The majority presented with open angle glaucoma but one individual presented with combined mechanism glaucoma. This individual appears to have

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<th>Table 1.</th>
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Ocular findings associated with each of the affected individuals.

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<th>Table 2.</th>
<th>Elbow flexion contracture</th>
<th>High arched palate</th>
<th>Pectus deformity</th>
<th>Prominent forehead</th>
<th>PIP joint prominence</th>
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Systemic findings associated with each of the affected individuals. The plus sign indicates that the finding is present in the individual and the minus sign indicates that the finding is missing in the individual.
an underlying primary open angle glaucoma compounded by the later development of anterior synechiae, possibly from intermittent pupillary block. Examination of the oldest individual was inconclusive due to complete corneal decompensation and the inability to obtain an adequate ocular examination. However, this individual had a history consistent with uncontrolled glaucoma and subsequent development of bilateral phthisis bulbi. Unfortunately, definitive records are not available. The remaining two individuals were the youngest in the family and do not currently have glaucoma; however, their affection status may change in the future.

Previous reports have demonstrated an association between Marfan syndrome and glaucoma [29,30]. Collagen is a major structural component throughout the anterior segment of the eye and fibrillin has been localized to the canal of Schlemm, the scleral spur [26], and the corneoscleral portion of the trabecular meshwork (TM) [31]. Therefore, one speculation on the development of glaucoma could involve changes in the structural support of these tissues leading to a decreased outflow facility and increased intraocular pressures. Abnormalities of the canal of Schlemm have been noted on histologic examination of Marfan eyes with displacement or discontinuities of this structure [32]. The elasticity of Schlemm’s canal is thought to influence aqueous outflow since collapse of the canal may impair fluid flow [33]. Therefore, a defective fibrillin 1 protein could potentially lead to increased elasticity and easier collapse of the canal of Schlemm. Furthermore, the scleral spur is thought to be involved in maintaining the normal architecture of the TM since processes from both the ciliary body and the corneoscleral portion of the TM insert into the scleral spur [34]. Moreover, one mechanism of the effect of low dose miotics in increasing the outflow facility of the eye is thought to involve ciliary muscle contraction against the scleral spur which subsequently increase the distraction of the trabecular meshwork [35]. Therefore, it is conceivable that a fibrillin mutation could affect the elasticity of the scleral spur and indirectly affect the trabecular meshwork architecture. Furthermore, another possible mechanism for the high glaucoma prevalence in this pedigree may be intermittent lens-induced pupillary block with subsequent development of anterior synechiae. However, despite numerous examinations, none of the family members were ever documented to be in actual pupillary block. Moreover, despite documented ocular pressures of 39 mmHg OD and 52 mmHg OS, the proband demonstrated gonioscopically open anterior chamber angles without evidence of pupillary block on examination.

Corneal changes typically associated with Marfan syndrome usually involve a flatter corneal curvature and astigmatism [36]. An association between Marfan syndrome and
mild corneal guttae formation has been reported in one series of 41 patients [37]. In our pedigree, the corneal guttae were not observed in the youngest affected members of this family. Guttae were, however, universally present in all affected adult family members and the oldest affected family member demonstrated frank corneal decompensation. The guttae observed could be indicative of an entirely separate process such as Fuch’s endothelial corneal dystrophy occurring in this family or it may represent an association between deranged fibrillin 1 production and the clinical appearance of guttace-like changes. Fibrillin 1 expression in descemet’s membrane has been demonstrated in corneal specimens from individuals with pseudophakic and aphakic bullous keratopathy [38]. To our knowledge, the presence of corneal guttae and the demonstration of progressively worse findings with advancing age is a feature not previously reported in Marfan syndrome.

In general, complete lens dislocation is a relatively rare occurrence in Marfan syndrome and to our knowledge this is the first reported family with bilateral juvenile onset lens dislocation. Additionally, this syndrome demonstrates a unique constellation of findings such as effective aphakia, increased axial length, associated central progressive corneal guttae formation and the development of glaucoma. This report serves to expand the phenotype of Marfan syndrome and its association with glaucoma and guttae needs further study.

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