Identification of novel CYP4V2 gene mutations in 92 Chinese families with Bietti’s crystalline corneoretinal dystrophy

Xiao Hong Meng,1,2 Hong Guo,3 Hai Wei Xu,1,2 Qi You Li,1,2 Xin Jin,1,4 Yun Bai,3 Shi Ying Li,1,2 Zheng Qin Yin1,2

1Southwest Hospital, Southwest Eye Hospital, Third Military Medical University, Chongqing, China; 2Key Lab of Visual Damage and Regeneration & Restoration of Chongqing, Chongqing, PR China; 3Department of Medical Genetics, Third Military Medical University, Chongqing, PR China; 4Department of Ophthalmology, Chinese PLA General Hospital, Beijing, PR China

Purpose: To characterize the spectrum of CYP4V2 gene mutations in 92 unrelated Chinese probands with Bietti’s crystalline dystrophy (BCD) and to describe the molecular and clinical characteristics of four novel CYP4V2 mutations associated with BCD.

Methods: All study participants underwent a complete ophthalmological examination. Mutational screening of CYP4V2 coding regions and flanking intron sequences was examined via directional Sanger sequencing, with allele separation confirmed by screening other family members. Subsequent in silico analysis of the mutational consequence on protein function was undertaken, with the impact of the novel mutation on pre-mRNA splicing examined via RT–PCR.

Results: Fifteen disease-causing variants were identified in 92 probands with BCD, including four novel mutations and eleven previously reported mutations. The most prevalent mutation was c.802_810del17insGC, which was detected in 69 unrelated families, with an allele frequency of 52.7% (97/184). Homozygosity was revealed in all 35 unrelated families, and compound heterozygosity was observed in 43 subjects. Four patients harbored four novel variants, with these mutations cosegregated within all affected individuals and were not found in unaffected family members and 100 unrelated controls. Transcriptional analysis of a novel splice mutation revealed altered RNA splicing. In silico analysis predicted that the missense variant, p.Tyr343Asp, disrupted the CYP4V2 surface electrostatic potential distribution and spatial conformation. Among the patients with four novel mutations, genotype did not always correlate with age at onset, disease course, or electroretinogram (ERG) changes, with phenotypic variations even noted within the same genotype.

Conclusions: The c.802_810del17insCG mutation was the most common mutation in the 92 Chinese probands with BCD examined. Four novel mutations were identified, contributing to the spectrum of CYP4V2 mutations associated with BCD, with no clear link established between disease phenotype and genotype.

Bietti’s crystalline corneoretinal dystrophy (BCD, MIM 210370) is an autosomal recessive retinal dystrophy that was first reported by Bietti in 1937 [1]. It is characterized by numerous tiny glistening yellow-white crystals scattered at the posterior pole of the retina, progressive atrophy of the RPE, and choroidal sclerosis; the majority of cases had similar crystals in the corneoscleral limbus. Patients with BCD usually present in the second or third decade, and they progress to legal blindness by the fifth or sixth decade of life [2].

The locus of the gene for BCD was mapped to 4q35, with mutations in the cytochrome P450, family 4, subfamily V, polypeptide 2 (CYP4V2, MIM 608614) gene associated with clinical phenotype. The CYP4V2 gene consists of 11 exons, encodes a 525 amino acid protein, and belongs to the CYP450 family. CYP4V2 is widely expressed in various tissues, including the human retina, RPE, lymphocytes, heart, brain, placenta, lung, liver, skeletal muscle, kidney, and pancreas, and has been thought to play a crucial role in fatty acid and corticosteroid metabolism [3,4].

BCD is relatively common in Eastern Asian populations, with CYP4V2 the only identified gene associated with the disease thus far. At present, 58 mutations have been described, which affect 47 amino acid positions within the protein [4-21]. All of these previously identified CYP4V2 mutations in patients with BCD were missense coding changes or insertions and/or deletions of one or a few amino acids. Among these mutations, the most common among Chinese patients with BCD include c.802_810del17insGC in exon 7, c.992A>C in exon 8, and c.1091–2A>G in an intronic, accounting for 83.3% of the mutant alleles [14]. To understand the distribution spectrum of these CYP4V2 mutations in Chinese patients with BCD, 92 unrelated probands were screened, and the molecular and clinical characteristics of novel CYP4V2 mutations were described.
METHODS

Recruitment of subjects: Ninety-two probands, clinically diagnosed with BCD, from unrelated families at the South-west Eye Hospital/Southwest Hospital, China, were recruited. Family members of the probands were also clinically examined, in addition to 100 normal controls who were referred for this study. The Ethics Review Board of the Southwest Hospital (Chongqing, China) approved all research protocols, which adhered to the tenets of the Declaration of Helsinki, with informed consent obtained from all participants. All probands underwent ophthalmological examinations including best-corrected visual acuity testing with the Snellen vision chart, slit-lamp biomicroscopy, and fundoscopy. Additionally, fundus photography was performed in 91 patients, automated perimetry (low vision model) in 18 patients, full-field electroretinography (FERG) in 82 patients, which was recorded according to the standards of the International Society for Clinical Electrophysiology of vision (ISCEV, 2008), and multifocal electroretinogram (mfERG) in 77 patients.

Mutation screening/detection: Blood samples were collected from 92 probands and their family members and 100 healthy controls, then preserved in freezers at -80 °C prior to use. Genomic DNA was extracted from whole blood using the

| Patient | Age/Gender | Age at onset of NB | Age at onset of DV | Refractive error (SE) | VA Snellen | Nucleotide change (Mutations) |
|---------|------------|--------------------|--------------------|-----------------------|------------|-----------------------------|
| 31      | 43/F       | 30                 | 11                 | RE: -10.5             | RE: 20/200 | c.283G>A, p.G95R+ c.1027T>G, p.Y343D* |
|         |            |                    |                    | LE: -10.5             | LE: 20/125 |                             |
| 33      | 61/F       | 55                 | 55                 | RE: -10.5             | RE: 20/400 | c.802–8_810del17insGC+ c.413+2T>G, del* |
|         |            |                    |                    | LE: HM                | LE: 20/125 |                             |
| 60      | 55/M       | 49                 | 44                 | RE: -2.75             | RE: 20/200 | c.31C>T, p.Q11X+ c.31C>T, p.Q11X* |
|         |            |                    |                    | LE: -2.75             | LE: 20/125 |                             |
| 86      | 44/M       | 41                 | 34                 | RE: 20/200            | RE: 20/200 | c.802–8_810del17insGC+ c.791 del T* |
|         |            |                    |                    |                       | LE: 20/50  |                             |

NB: night blindness. DV: decrease of visual acuity. RE: right eye. LE: left eye. * Novel mutation.
Tiangen blood kit (Tiangen Biltech, Beijing, China) following the manufacturer’s standard sequencing protocols. Genomic DNA was extracted from whole blood using the Tiangen blood kit (Tiangen Biltech) according to the manufacturer’s protocols. All CYP4V2 coding exons, including intron–exon boundaries, were amplified in the 92 probands samples via PCR using primers previously described by Li [12]. The PCR products were subsequently purified with a TIANGen Midi Purification Kit (Tiangen Biltech), sequenced with an ABI BigDye Terminator Cycle Sequencing kit v3.1 (ABI Applied Biosystems, Foster City, CA), and DNA sequences analyzed using the vector NTI 10.3 software package. CYP4V2 orthologs were also investigated with the NCBI HomoloGene database.

Total RNA was extracted from white blood cells using TRIzol Reagent (Life Technologies, Carlsbad, CA) according to the manufacturer’s protocol. An RNA PCR kit (TaKaRa, Dalian, China) was used to synthesize cDNA from 2 μg of RNA according to the manufacturer’s protocols. During the RT–PCR reaction, cDNA was amplified using the primers: RT1-F: 5′-GAA GCC GGA CGG GCG AGA AT-3′, RT1-R: 5′-CTC GTG CGC CAT TTG TTT-3′ to generate CYP4V2 fragments.

**Functional significance prediction:** To illustrate the effects of novel CYP4V2 mutations, the conservation of p.Tyr343, p.Gln11, and p.Phe264 was analyzed among eight species, the molecular structures of novel missense mutation was analyzed in silico, and pre-mRNA splice-site mutations were confirmed with reverse transcription-PCR (RT-PCR).

The crystal structure of CYP4V2 was modeled based on the crystal structure of CYP46A1, a member of the cytochrome P450 superfamily (PDB ID: 3mdm) and an enzyme that initiates the major pathway of cholesterol elimination from the brain, with an amino acid sequence homology of 25%. A molecular model of CYP4V2 was constructed with the SWISS-MODEL server, with structural graphics and visualization based on the Swiss-Pdb Viewer 4.04.

**RESULTS**

**Clinical findings:** Of the 92 probands, 49 were male and 43 were female, with five consanguineous individuals in the cohort. The affected individuals presented ages of initial
Symptoms of night blindness from 17 to 55, with most individuals in their 30s. Fundus observations revealed typical BCD changes in all 92 probands, including numerous small yellow-white crystalline spots distributed on the macula, posterior poles, and peripheral retina, with the number of crystalline spots varying significantly among patients. Examination of all probands revealed 89 patients (96.7%) with RPE atrophy (age range from 24 to 61), ten patients (10.9%) with severe choroidal sclerosis (age range from 25 to 61), 39 patients (42.4%) with crystals in the limbus cornea (age range from 31 to 62), and 70 patients (76.1%) with vitreous crystals (age range from 20 to 62). Visual acuity varied markedly from light perception (LP) to 20/20, and 61/92 patients (67.8%) showed low vision (best-corrected visual acuity worse than 0.5 logMAR but equal or better than 1.3 logMAR in the better eye, which equals the Snellen chart 20/63). Low visual peripheral (LVP) analysis further showed 14 patients (22 eyes) with irregular visual field defects (mean sensitivity, MS, 10.1 to 26.9) and nine patients (14 eyes) with retained visual islands (MS 2.6 to 10.2). For the ERG recordings, 38 patients (41.3%, 74 eyes) had a non-recordable rod response, only three patients (3.3%, five eyes) showed a rod dominant ERG, and 43 patients (46.7%, 85 eyes) showed a cone dominant ERG. Furthermore, mfERG recordings showed 58 patients (60.9%, 116 eyes) had severe reductions in the response amplitudes of the central and peripheral macula, seven patients (7.6%, 14 eyes) had non-recordable mfERG.

### Table 2. CYP4V2 Mutations Identified in 92 Families with BCD.

| Exon | DNA Change       | Amino acid change | Type of nucleotide change | Allele frequency in |
|------|------------------|-------------------|---------------------------|---------------------|
|      |                  |                   | hom/heter                 | 100 Controls        | 92 probands | Note |
| 1    | c.31C>T          | p.Q11X            |                           | 1/0                 | 0/200       | 2/184   | novel |
| 1    | c.64C>G          | p.L22V            |                           | 1/4                 | ND          | 6/184   | Reported |
| 2    | c.215–2A>G       | Splicing acceptor |                           | 2/0                 | ND          | 4/184   | Reported |
| 2    | c.219T>A         | p.F73L            |                           | 0/3                 | 0/200       | 3/184   | Reported |
| 2    | c.283G>A         | p.G95R            |                           | 0/5                 | ND          | 5/184   | Reported |
| 3    | c.413+2T>G       | Splicing acceptor |                           | 0/1                 | 0/200       | 1/184   | novel |
| 6    | c.732G>A         | p.W244X           |                           | 0/1                 | ND          | 1/184   | Reported |
| 6    | c.791delT        | delet             |                           | 0/1                 | 0/200       | 1/184   | novel |
| 7    | c.802–8_810del17insGC | Splicing acceptor |                           | 28/41               | ND          | 97/184  | Reported |
| 7    | c.958C>T         | p.R320X           |                           | 0/1                 | ND          | 1/184   | Reported |
| 8    | c.992A>C         | p.H331P           |                           | 0/22                | ND          | 22/184  | Reported |
| 8    | c.1027T>G        | p.Y343D           |                           | 0/1                 | 0/200       | 1/184   | novel |
| 8    | c.1061–1062insA  | Ins               |                           | 0/2                 | ND          | 2/184   | Reported |
| 9    | c.1091–2A>G      | Splicing acceptor |                           | 3/11                | ND          | 17/184  | Reported |
| 9    | c.1187C>T        | p.P396L           |                           | 0/2                 | ND          | 2/184   | Reported |
|      | Total            |                   |                           | 35/95              | 165/184     |         |
recordings, and only 12 patients (10.9%, 23 eyes) with BCD had mild reductions of mfERG.

The clinical characteristics of the four patients with novel mutations were analyzed, with the age at onset of first symptoms of night blindness and decreased visual acuity ranging from 11 to 55 (Table 1). Visual acuities ranged from 20/50 to hand movement (HM), and all patients had characteristic numerous small retinal crystalline deposits, varying degrees of RPE atrophy, and choroidal sclerosis in the posterior pole. Fundus revealed patient 31, patient 85, and patient 86 had serious choroidal sclerosis (Figure 1). Slit-lamp examination showed two patients had peripheral corneal crystalline deposits. FERG responses were extinct in three patients (patient 31, patient 60, patient 86; Figure 2). Multifocal ERG response was severely reduced in four patients.

Mutation analysis: Following sequence analysis of the CYP4V2 coding and adjacent intronic regions, homozygous (35 families), compound heterozygous (43 families), and heterozygous mutations (nine families) were detected, with no CYP4V2 mutation detected in five families. Fifteen mutations were identified in total, with four mutations identified as novel (Table 2).

The c.802_810del17insGC mutation was detected in 69 unrelated families, with an allele frequency of 52.7% (97/184), while the c.992A>G mutation accounted for 11.9% (22/184), and the c.1091–2A>G accounted for 9.8% (17/184). Collectively, these three common mutations comprise 73.37% of the mutations of CYP4V2, with c.802_810del17insGC the most prevalent mutation in 92 Chinese families with BCD. The results further demonstrate that mutation c.802_810del17insGC is one of the most common mutations in Chinese populations.

Four non-consanguineous patients had four novel mutations, which have not been previously reported (Figure 3). The underlying novel variations revealed three kinds of compound heterozygosity and one homozygous mutation in the four patients. The novel mutations were cosegregated within all affected individuals and were not found in the 100 Chinese control individuals. A missense mutation, c.1027T>G, was identified in patient 31 (Figure 4A,B), which

![Figure 4](http://www.molvis.org/molvis/v20/1806>)

Figure 4. Genomic DNA and cDNA of the CYP4V2 gene in four novel mutations. A: The heterozygous mutant single base-pair change (c.1027T>G, p.Y343D). B: Control (wild type, WT). C: The homozygous nonsense mutant (p.Q11). D: Control (WT). E: The heterozygous change (c.791del T). F: Control (WT). G: DNA-MT of the heterozygous splice change (c.413+2T>G). H: Control (WT). I: cDNA sequence, from the sense primer, for the control (WT). J: The cDNA of the heterozygous splice change (c.413+2T>G). The deletion is indicated with an arrow and black frame.
resulted in a tyrosine to aspartic acid change (p.Tyr343Asp).
In patient 60, a nonsense mutation (c.31C>T) that converts a glutamine (CAG) into a premature termination codon (TAG) at amino acid position 11 (p.Gln11X) in exon 1 was identified (Figure 4C,D), resulting in the conversion of a 525-amino acid sequence to an 11-amino acid (MAGLWLGLVWQ) sequence.
A heterozygous deletion, c.791delT, which causes the deletion of an amino acid at position 264, resulting in an in-frame shift, was identified in patient 86 (Figure 4E,F). In patient 33, one novel heterozygous splice-site mutation, c.413+2T>G, was detected in the 3′- acceptor in intron 3, resulting in a GTA to GTA (Figure 4G,H). Furthermore, RT–PCR showed that the cDNA nucleotide sequence derived from the mutant transcript resulted in a truncation of exon 3, resulting in a 87-bp/29-amino acid deletion (Figure 4I,J).

Conservation of novel mutations in CYP4V2: CYP4V2 encodes a 525-amino acid protein that belongs to a member of the CYP450 family and is widely expressed in human tissues including the sensory retina and the RPE. A CYP4V2 ortholog investigation revealed that p.F264 was highly conserved among eight species although p.Q11 and p.Y343 were not completely conserved across all eight species (Figure 5).

Figure 5. The alignment of novel mutations of the CYP4V2 gene with the corresponding segments in eight species. Amino acids marked in the column are highly conserved among all shown species in F264, while Q11 is conserved among five shown species and Y343 is conserved among seven shown species. Background color illustration: transparent=non-similar; light blue=conserved; yellow=identical.

Figure 6. Superimposed wild-type and mutant CYP4V2 and electrostatic potential differences between WT and Y343D. A: Wild-type CYP4V2 (72–523; pink) and CYP4V2 (72–523) Y343D (blue) is shown on the right. The typically disturbed secondary structures are magnified for detail. B: Blue denotes a positive charge, and red denotes a negative charge. The arrows indicate the charge distribution change region on the molecular surface in the mutants and the wild type (WT).
Mutant structure modeling of one missense mutation: Mutant molecular model showed that p.Tyr343Asp affected the alpha spiral structure on the surface region of the protein (Figure 6A), and disrupting the surface electrostatic potential distribution and the spatial conformation of CYP4V2 (Figure 6B).

DISCUSSION

In this study, a total of 15 CYP4V2 mutations were identified from the 92 probands with BCD and 92 unrelated families of Chinese ethnicity; four were novel pathogenic variants. Thus far, this is one of the largest BCD-associated CYP4V2 mutational screenings in a Chinese population. Among the 15 mutations, the nucleotide deletion/insertion c.802–810del17insGC was the most frequently occurring pathogenic mutation, which could be a common founder effect contributing to the high prevalence in the Chinese population. The c.992A>G and c.1091–2A>G mutations were also common in this study, with these findings in agreement with previous studies [4,8,12,14].

In our cohort, four novel mutations were identified in the CYP4V2 gene, with targeted mutational analysis of family members confirming that these four novel mutations, p.Tyr343Asp, c.413+2T>G, c.791delT, and p.Gln11X, segregated with the BCD phenotypes and were absent from a panel of 100 normal unrelated controls. These four novel mutations may affect the structure and function of CYP4V2, such as the c.413+2T>G mutation that was located in a splice site and led to a protein truncation. This deletion of 87 bp, resulting in the deletion of 29 amino acids, must affect the structural stability and function of CYP4V2. Second, a novel nonsense mutation (p.Gln11X) in exon 1 causes premature termination and eliminates a large portion of the CYP4V2 structure. Third, due to a frameshift deletion mutation, c.791delT in exon 6, the amino acid sequence was altered to create a false sequence, resulting in an abnormal protein structure and a nonfunctioning CYP4V2. For the novel heterozygous missense mutation, in silico analysis revealed that p.Tyr343, amino acid substitutions not only changed the local random coil structure on the surface of CYP4V2 but also disrupted the surface electrostatic potential distribution and space conformation. Although mutation, p.Tyr343, did not completely at a highly conserved site in cytochrome P450, it is a low frequency mutation in CYP4V2, and database for nonsynonymous SNPs’ functional predictions (dbNSFP) prediction showed that p.Tyr343 is a damaging and disease-causing mutation. However, further investigation is needed to elucidate the pathogenic mechanisms of the novel missense mutations.

Earlier reports have suggested that different CYP4V2 mutations could cause divergent BCD phenotypes [9,10,12,17,18]. In this study, we described the genotype-phenotype correlations for four patients with novel mutations. The phenotypes consisted of varying degrees of crystalline deposits, choroidal sclerosis, RPE atrophy, age of onset, disease course, and ERG changes, with severity not always associated with genotype. Patient 60, who had a deleterious homozygous novel nonsense mutation (p.Q11X), did not exhibit earlier disorder symptom and disease severity relative to the patients with heterozygous missense mutations or splice mutations.

Patient 33, who harbored a deletion/insertion mutation (c.802–8_810del17insGC) and a novel splice mutation (c.413+2T>G), exhibited a late age of onset, rapid disease progression, and severe vision loss compared with other patients with splice mutations in our cohort. It was also discovered that ERG response in these patients did not seem to be associated with the type of mutation. Among the four patients with novel mutations, FERG was extinct in four patients, and the mfERG response was severely reduced in four patients with different mutations, which is different from previous reports [12]. Moreover, age of onset did not always correlate with genotype severity. For example, patient 31, who had compound heterozygous missense mutations (G95R and Y343D), exhibited early onset, had a relatively slow disease progression, and presented with similar severe morphofunctional changes as those seen in patient 60 (Q11X), who exhibited late age of onset and fast disease progression. Furthermore, the age of onset varied among patients with the same mutations, further supporting the high variability between the BCD genotype and phenotype. This suggests that the clinical phenotype severity in patients with BCD cannot be determined by genotype; thus, other factors such as gene modifications, gene regulation, and environmental factors may influence the interchange between pathogenicity and CYP4V2 mutations.

Interestingly, this study discovered seven patients (8.9%) with BCD exhibited high myopia, which has not been reported in other Chinese families with BCD. There was no evidence that this myopia directly accelerated BCD progress in our primary study.

We observed that patients with high myopia appeared to have only poorer best-corrected visual acuity and didn’t show more affected retinal functioning based on ERG response. Due to the lack of continuous observation of retinal function changes of continuous and binocular disparity, it is difficult to confirm that the ERG responses in these patients with BCD associated with high pathological axial myopia.
showed additional decreases. In fact, the amplitude of the a wave and the b wave on ERG decreased in the axial high myopia eyes (ocular axial length is more than 26 mm, equivalent diopter (D) greater than 6.25D). Retinal degeneration diseases combined with high myopia are rare, with some studies suggesting a compounding effect, but currently, there is not enough evidence to prove a correlation with the retinitis pigmentosa (RP) clinical phenotype [22-25]. Further observation is needed to determine if the combined ocular complications in BCD aggravate retinal function.

In conclusion, the c.802_810del17insCG mutation was found to be the most common mutation in 92 Chinese probands with BCD. Four novel mutations were identified, contributing to the spectrum of CYP4V2 mutations associated with BCD, with no clear link established between disease phenotype and genotype.

ACKNOWLEDGMENTS

The authors are grateful to all patients and their family members for their participation in this study. The authors have no proprietary or commercial interest in any materials discussed. This work was supported by the National Basic Research Program of China (No 2013CB967002), and the National Nature Science Foundation of China (Grant No, 81130017)

REFERENCES

1. Bietti G. Ueber familiare Vorkommen von ‘Retinitis punctata albinescens’ (verbunden mit ‘Dystrophia marginalis crystallinea corneae’) Glitzern des Glaskoerpers und anderen degenerativen Augenveranderungen. Klin Mbl Augenheilk. 1937; 99:739-57.

2. Kaiser-Kupfer MI, Chan CC, Markello TC, Crawford MA, Caruso RC, Csaky KG, Guo J, Gahl WA. Clinical biochemical and pathologic correlations in Bietti’s crystalline dystrophy. Am J Ophthalmol 1994; 118:569-82. [PMID: 7977570].

3. Jiao X, Munier FL, Iwata F, Hayakawa M, Kanai A, Lee J, Schorderet DF, Chen MS, Kaiser-Kupfer M, Hejtmancik JF. Genetic linkage of Bietti crystallin corneoretinal dystrophy to chromosome 4q35. Am J Hum Genet 2000; 67:1309-13. [PMID: 11001583].

4. Li A, Jiao X, Munier FL, Schorderet DF, Yao W, Iwata F, Hayakawa M, Kanai A, Shy Chen M, Alan Lewis R, Heck-enlively J, Weleber RG, Traboulsi EI, Zhang Q, Xiao X, Kaiser-Kupfer M, Sergeev YV, Hejtmancik JF. Bietti crystalline corneoretinal dystrophy is caused by mutations in the novel gene CYP4V2. Am J Hum Genet 2004; 74:817-26. [PMID: 15042513].

5. Gekka T, Hayashi T, Takeuchi T, Goto-Omoto S, Kitahara K. CYP4V2 mutations in two Japanese patients with Bietti’s crystalline dystrophy. Ophthalmic Res 2005; 37:262-9. [PMID: 16088246].

6. Lee KY, Koh AH, Aung T, Yong VH, Yeung K, Ang CL, Vithana EN. Characterization of Bietti crystalline dystrophy patients with CYP4V2 mutations. Invest Ophthalmol Vis Sci 2005; 46:3812-6. [PMID: 16186368].

7. Lin J, Nishiguchi KM, Nakamura M, Dryja TP, Berson EL, Miyake Y. Recessive mutations in the CYP4V2 gene in East Asian and Middle Eastern patients with Bietti crystalline corneoretinal dystrophy. J Med Genet 2005; 42:e38-[PMID: 15937078].

8. Shan M, Dong B, Zhao X, Wang J, Li G, Yang Y, Li Y. Novel mutations in the CYP4V2 gene associated with Bietti crystalline corneoretinal dystrophy. Mol Vis 2005; 11:738-43. [PMID: 15860296].

9. Wada Y, Itabashi T, Sato H, Kawamura M, Tada A, Tamai M. Screening for mutations in CYP4V2 gene in Japanese patients with Bietti’s crystalline corneoretinal dystrophy. Am J Ophthalmol 2005; 139:894-9. [PMID: 15860296].

10. Jin ZB, Ito S, Saito Y, Inoue Y, Yanagi Y, Nao-i N. Clinical and molecular findings in three Japanese patients with crystalline retinopathy. Jpn J Ophthalmol 2006; 50:426-31. [PMID: 17013694].

11. Nakamura M, Lin J, Nishiguchi K, Kondo M, Sugita J, Miyake Y. Bietti crystalline corneoretinal dystrophy associated with CYP4V2 gene mutations. Adv Exp Med Biol 2006; 572:49-53. [PMID: 17013694].

12. Lai TY, Ng TK, Tam PO, Yam GH, Ngai JW, Chan WM, Liu DT, Lam DS, Pang CP. Genotype phenotype analysis of Bietti’s crystalline dystrophy in patients with CYP4V2 mutations. Invest Ophthalmol Vis Sci 2007; 48:5212-20. [PMID: 17962476].

13. Zenteno JC, Ayala-Ramirez R, Graue-Wiechers F. Novel CYP4V2 gene mutation in a Mexican patient with Bietti’s crystalline corneoretinal dystrophy. Curr Eye Res 2008; 33:313-8. [PMID: 18398705].

14. Xiao X, Mai G, Li S, Guo X, Zhang Q. Identification of CYP4V2 mutation in 21 families and overview of mutation spectrum in Bietti crystalline corneoretinal dystrophy. Biochem Biophys Res Commun 2011; 409:181-6. [PMID: 21565171].

15. Yokoi Y, Sato K, Aoyagi H, Takahashi Y, Yamagami M, Nakazawa M. A Novel Compound Heterozygous Mutation in the CYP4V2 Gene in a Japanese Patient with Bietti’s Crystalline Corneoretinal Dystrophy. Case Rep Ophthalmol 2011; 2:296-301. [PMID: 22087103].

16. Mamatha G, Umashankar V, Kasinathan N, Krishnan T, Sathyabaraathri R, Karthiyyinini T, Amali J, Rao C, Madhavan J. Molecular screening of the CYP4V2 gene in Bietti crystalline dystrophy that is associated with choroidal neovascularization. Mol Vis 2011; 17:1970-7. [PMID: 21850171].

17. Haddad NM, Waked N, Bejjani R, Khoueir Z, Chouery E, Corbani S, Megarbane A. Clinical and molecular findings in three Lebanese families with Bietti crystalline dystrophy:
report on a novel mutation. Mol Vis 2012; 18:1182-8. [PMID: 22605929].

18. Rossi S, Testa F, Li A, Yaylacioglu F, Gesualdo C, Hejtmancik JF, Simonelli F. Clinical and genetic features in Italian Bietti crystalline dystrophy patients. Br J Ophthalmol 2013; 97:174-9. [PMID: 23221965].

19. García-García GP, Lopez-Garrido MP, Martinez-Rubio M, Moya-Moya MA, Belmonte-Martinez J, Escribano J. Genotype-phenotype analysis of Bietti crystalline dystrophy in a family with the CYP4V2 Ile111Thr mutation. Cornea 2013; 32:1002-8. [PMID: 23538635].

20. Halford S, Liew G, Mackay DS, Sergouniotis PI, Holt R, Broadgate S, Volpi EV, Ocala L, Robson AG, Holder GE, Moore AT, Michaelides M, Webster AR. Detailed phenotypic and genotypic characterization of Bietti crystalline dystrophy. Ophthalmology 2014; 121:1174-84. [PMID: 24480711].

21. Yin H, Jin C, Fang X, Miao Q, Zhao Y, Chen Z, Su Z, Ye P, Wang Y, Yin J. Molecular analysis and phenotypic study in 14 Chinese families with Bietti crystalline dystrophy. PLoS ONE 2014; 9:e94960-[PMID: 24739949].

22. den Hollander AI, McGee TL, Ziviello C, Banfi S, Dryja TP, Gonzalez-Fernandez F, Ghosh D, Berson EL. A homozygous missense mutation in the IRBP gene (RBP3) associated with autosomal recessive retinitis pigmentosa. Invest Ophthalmol Vis Sci 2009; 50:1864-72. [PMID: 19074801].

23. Sheng X, Li Z, Zhang X, Wang J, Ren H, Sun Y, Meng R, Rong W, Zhuang W. A novel mutation in retinitis pigmentosa GTPase regulator gene with a distinctive retinitis pigmentosa phenotype in a Chinese family. Mol Vis 2010; 16:1620-8. [PMID: 20806050].

24. Mäntyjärvi M, Katajakunnas M, Vanttinen S. High myopia with cone dysfunction. Acta Ophthalmol (Copenh) 1991; 69:155-61. [PMID: 1872133].

25. Vaclavik V, Gaillard M-C, Tiab L, Schorderet DF, Munier FL. Variable phenotypic expressivity in a Swiss family with autosomal dominant retinitis pigmentosa due to a T494M mutation in the PRPF3 gene. Mol Vis 2010; 16:467-75. [PMID: 20309403].

Articles are provided courtesy of Emory University and the Zhongshan Ophthalmic Center, Sun Yat-sen University, P.R. China. The print version of this article was created on 31 December 2014. This reflects all typographical corrections and errata to the article through that date. Details of any changes may be found in the online version of the article.