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Novel Compound Heterozygous *CBS* Mutations Cause Homocystinuria in a Han Chinese Family

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The *cystathionine β-synthase* (*CBS*) gene has been shown to be related to homocystinuria. This study was aimed to detect the mutations in *CBS* in a Han Chinese family with homocystinuria. A four-generation family from Shandong Province of China was recruited in this study. All available members of the family underwent comprehensive medical examinations. Genomic DNA was collected from peripheral blood of all the participants. The coding sequence of *CBS* was amplified by polymerase chain reaction (PCR), followed by direct DNA sequencing. Among all the family members, three affected individuals showed typical clinical features of homocystinuria. Two novel compound heterozygous mutations in the *CBS* gene, c.407T > C (p. L136P) and c.473C > T (p.A158V), were identified by sequencing analysis in this family. Both of the two missense mutations were detected in the three patients. Other available normal individuals, including the patients' parents, grand parents, her younger sister and brother in this family either carried one of the two mutations, or none. In addition, the two mutations were not found in 600 ethnically matched normal controls. This study provides a mutation spectrum of *CBS* resulting in homocystinuriain a Chinese population, which may shed light on the molecular pathogenesis and clinical diagnosis of *CBS*-associated homocystinuria.

Homocystinuria, most commonly caused by cystathionine β-synthase (*CBS*) deficiency, is an autosomal recessive disorder of sulfur amino acid metabolism. *CBS* protein is a pyridoxal 5′ phosphate dependent enzyme and catalyzes the condensation of homocysteine with homocysteine and serine to form cystathionine[1]. Biochemically, this disorder is characterized by elevated plasma concentrations of homocysteine and methionine, increased excretion of homocysteine in urine and decreased levels of cystathionine and cysteine in body fluids[2]. Patients with homocystinuria often display different symptoms,including ocular anomalies (severe myopia and ectopialentis), skeletal deformities (osteoporosis, scoliosis and Marfanoid habitus), vascular thrombosis and ischemia, disorder of central nervous system (mental retardation, convulsions and psychiatric disturbances) and other manifestations[3].

It has been reported that homocystinuria due to *CBS* deficiency is caused by mutations in the *CBS* gene[4]. *CBS* mutations could lead to the disruption of enzyme activity which consequently results in increased levels of homocysteine, a potentially toxic amino acid responsible for patients with homocystinuria. So far, more than 150 mutations in the *CBS* gene have been identified (http://cbs.lf1.cuni.cz/mutations.php) in different ethnic population[5–12]. Although many mutations have been described in the *CBS* gene from homocystinuric patients and this disease has been well characterized in other populations, the range of clinical presentations and spectrum of *CBS* mutations in Han Chinese patients remained largely uninvestigated.

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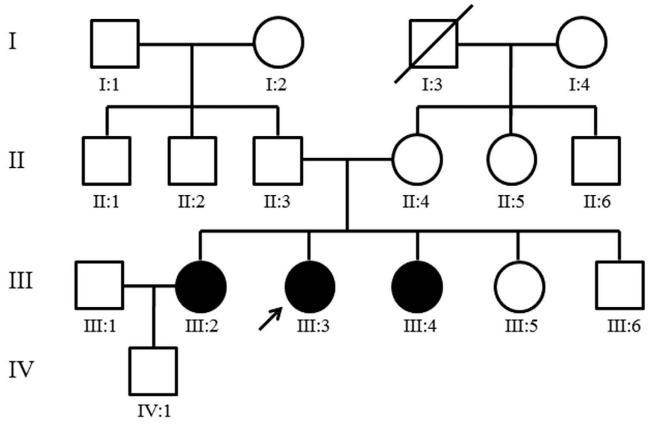

Figure 1. Pedigree of the family with homocystinuria. Solid symbols indicated affected individuals, and open symbols indicate unaffected individuals. Arrow indicates the proband.

Patient number	III:2	III:3	III:4
Age (Year)/Sex[1]	28/F	26/F	25/F
Onset age (Year)	5	7	5
Eye involvement	Biocular lens dislocation (were extracted 11 years ago); Myopia;Exotropia; Corneal staphyloma; Retinal detachment	Biocular lens dislocation (were extracted 11 years ago); Myopia; Exotropia; Corneal staphyloma	Biocular lens dislocation (were extracted 8 years ago); Myopia; Exotropia; Corneal staphyloma
Skeletal system	Kyphoscoliosis	Arachnodactylies	Arachnodactylies; Kyphoscoliosis; Mild pectuscarinatum
IQ[2]	Mental retardation; Dysarthria	Mental retardation; Dysarthria	Mental retardation; Dysarthria
Other signs	Pyramidal signs;Ataxia;Unstable gait;Brain atrophy; Malar flush	Pyramidal signs;Ataxia; Unstable gait;Brain atrophy; Malar flush	Pyramidal signs; Ataxia; Unstable gait;Brain atrophy; Malar flush
tHcy[3]	103	97	86
P-Met[4]	345	287	295

Table 1. Clinical data of affected members in this family with homocystinuria. [1]F, female; M, male. [2]IQ, intelligence quotient (assessed at presentation, various tests were used). [3]tHcy, plasma total homocysteine level at presentation (μ mol/L), reference range 5–15 μ mol/L. [4]P-Met, plasma methionine level at presentation (μ mol/L), reference range 20–40 μ mol/L.

In this study, we characterized the clinical manifestations and investigated the molecular basis of a Han Chinese family with homocystinuria, to expand the *CBS* mutation spectrum of the homocystinuric patients from China.

Materials and Methods

Subjects. This family with homocystinuria, including 16 members, was recruited from Shandong Provincial Hospital Affiliated to Shandong University (Fig. 1). Three of the family members were diagnosed as homocystinuria by biochemical profiles (grossly increased serum homocysteine and methionine) and by critical complications, such as dislocated lens, mental retardation and skeletal deformities. Their clinical information is summarized in Table 1.This study was conducted in accordance with the tenets of the Declaration of Helsinki and approved by the Institutional Review Boards of Hospital of University of Electronic Science and Technology of China & Sichuan Provincial People's Hospital, and Shandong Provincial Hospital Affiliated to Shandong University. Written informed consents were obtained from the family prior to the study.Unrelated healthy control subjects were recruited from the Hospital of University of Electronic Science and Technology of China and Sichuan Provincial People's Hospital. These controls are all Han Chinese, and subjects were excluded from the study if they had any of the symptoms, including ocular anomalies, skeletal deformities, vascular thrombosis and disorder of central nervous system.

DNA Extraction. All genomic DNA was extracted from peripheral blood using a blood DNA extraction kit (QIAamp DNA Blood Midi Kit; Qiagen, Germany) according to the manufacturer's protocol. DNA samples were stored at −20 °C until used. DNA integrity was evaluated by 1% agarose gel electrophoresis.

Mutation screening. The method for mutation screening was performed as described previously[13]. Besides the variants in *CBS*, mutations in the *MTHFR* gene also have been reported to cause homocystinuria[14,15]. Therefore, the coding sequences of *CBS* (NM_000071.2) and *MTHFR*(NM_005957.4) were amplified by polymerase chain reaction (PCR) using a MyCycler thermo cycler (Bio-Rad, Hercules, CA). We did not detect any mutation in the *MTHFR* gene in this family (data not shown), thus it was excluded to cause homocystinuria in this study. Sequencing primers from flanking sequence of each exon of the *CBS* gene were designed by using the Primer 5.0

Primer Name	Primer Sequence(5′–3′)	Product Size (bp)	Annealing Temperature (°C)
CBS 1F	CTCTCTCCTTGCTTTGCCAG	469	59
CBS 1R	CTGAGCATCCACTGTCTTGC		
CBS 2F	ATGTGTGTTTCAGGCGTGTG	453	59
CBS 2R	GCCACTCATTAACCAGCGAG		
CBS 3F	GGGGAGAAGCTCTGATAGGC	516	59
CBS 3R	CCGAATGCTGGTCAAAGGAA		
CBS 4&5&6F	CCATGTTGGGCAATTTTGGA	772	59
CBS 4&5&6R	AGCATTTCACAGAGGGAACA		
CBS 7F	CTTTCACAGACCAAGGGCAG	400	65
CBS 7R	TCTTCCCAAACACCTCCCAG		
CBS 8F	TTGGGTTTCTCATCCTGCCT	448	59
CBS 8R	GACCTTCGAGACCAGCTTCT		
CBS 9F	CTGTCTGCAAAACGTGTTGG	400	59
CBS 9R	CGCAGTGACACTCCTCAGAA		
CBS 10F	GCACAAGGAAGAAGCCGATG	366	59
CBS 10R	GTGAGAGGCATCCAGGGAAG		
CBS 11&12F	GCATGCTCACACACGCTT	819	59
CBS 11&12R	TGCCCTGAACGTCTGTATGA		
CBS 13F	CGAGGACATGTCTGACAGCA	975	65
CBS 13R	GAGTACTCTGGCACCCTCTG		
CBS 14F	CTGCCCAAACCTAGGAGTGA	432	59
CBS 14R	ACTGGGTGTCACTGAAGGTC		
CBS 15F	GGAGTCTGAGGCACGAGAAT	432	65
CBS 15R	GAAAGCGAAGGAGAAGTGGG		

Table 2. Primers used for mutation screening in *CBS* gene. F: Forward primer; R: Reverse primer; bp: Base pair.

(Table 2). Amplification reaction was performed by the PCR reaction (10 μL final volume) containing 50 ng of genomic DNA, 1 μL of each primer (10 pmol/μL), 1 μL of 10 buffer (Takara Bio Inc., Shiga, Japan), 0.8 μL of deoxyribonucleotide triphosphates (2 mmol/L; Takara Bio Inc.), 0.4 μL $MgCl_2$ (2.5 mmol/L; Takara Bio Inc.), and 0.1 μL of ExTaq polymerase (5 U/μL; Takara Bio Inc.). Amplified PCR products were purified with spin columns (QIAquick, Qiagen, Valencia, CA) and sequenced directly (BigDye Terminators Sequencing Kit; Applied Biosytems) in both directions with an automated genetic analysis system (ABI 3130 Genetic Analyzer, CA, USA).

Multiple sequence alignment of the human *CBS* protein was performed along with other *CBS* protein across different species, to check for the conservation of the residues. The possible damaging effects of the 2 mutations on the structure and function of *CBS* were predicted using SIFT (http://sift.jcvi.org) and PolyPhen-2 (http://genetics.bwh.harvard.edu/pph2/).

Results

Clinical findings. A four-generation family from Shandong Province of China was recruited in this study (Fig. 1). There are three affected individuals (III:2, III:3 and III:4), who showed typical clinical symptoms of homocystinuria among all the family members. The proband (III:3), as well as her two affected sisters(III:2 and III:4) exhibited similar clinical features, such as various reduced visual acuities with a bilateral lens dislocation, myopia, glaucoma, skeletal deformities and mental retardation(Table 1). All the patients also have elevated plasma homocysteine and methionine levels, compared to all the normal individuals of this family (±13.6 μmol/L for homocysteine and ±24.7 μmol/L for methionine, respectively). The parents, the grand parents and other relatives of the three affected individuals had no homocystinuric symptoms, exhibiting a pattern of recessive inheritance in this family.

Mutation screening of *CBS* in homocystinuria. Sequencing analysis of the *CBS* gene revealed novel compound heterozygous mutations, c.407T > C(p. L136P) and c.473C > T (p.A158V) (Fig. 2). They located in the coding sequence at nucleotide 407 of exon 3 and at nucleotide 473 of exon 4, respectively (Fig. 3). Both the two missense mutations were present in the three affected subjects (Table 3). The parents of the three patients were unaffected carriers with c.473C > T(father) and c.407T > C(mother) mutations, showing complete co-segregation of the mutations with the disease phenotype. Other available normal individuals in this family either carried one of the two mutations, or none. In addition, neither of the two missense heterozygous mutations was detected in 600 ethnically matched normal controls.

Comparative amino acid sequence alignment of other *CBS* protein across different species revealed that the two novel mutations occurred at highly conserved positions (Fig. 4). Both of the two novel mutations could result in substitutions of amino acid in the *CBS* protein, and were predicted to be damaging by SIFT and Polyohen 2 (Table 3). The c.407T > C mutation is a T-C transition, converting Leucine (L) to Proline (P) at amino acid 136

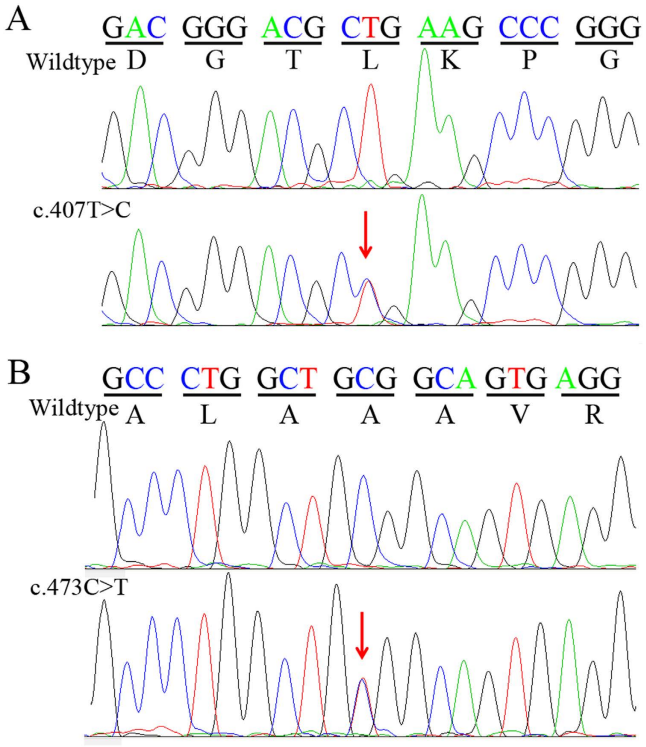

Figure 2. Direct sequencing results of *CBS* mutations. Direct sequencing identified two novel compound heterozygous mutations, (**A**) c.407T > C(p. L136P) and (**B**) c.473C > T (p.A158V)(indicated by red arrow).

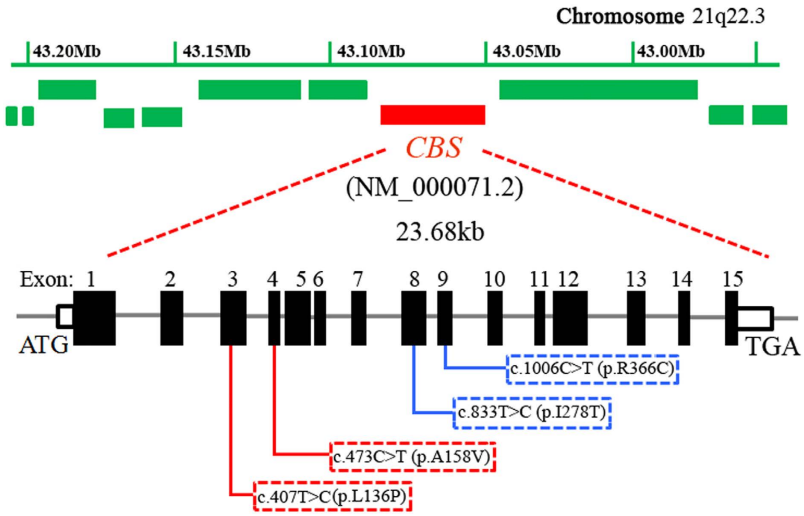

Figure 3. Mutations in the *CBS* gene identified in Chinese homocystinuric patients. The boxed mutations in red were newly found in this study and the mutations in blue box were identified in a Hong Kong homocystinuric patient.

(p. L136P); another mutation is a C-T transition (c.407T > C), leading to substitution of Alanine (A) toValine (V) at codon 158 (p.A158V, Fig. 2).

Discussion

Homocystinuria is the most common inborn disorder of sulfur amino acid metabolism. *CBS* deficiency, a main factor causing homocystinuria, is an autosomal recessively inherited genetic defect. Since the first mutation in the human *CBS* gene reported by Kozich and Kraus in 1992[4], many *CBS* mutations in homocystinuric patients from various populations worldwide have been identified. The present study identified novel compound heterozygous mutations, c.407T > C (p. L136P) and c.473C > T (p.A158V), in a Han Chinese family with homocystinuria and this result expands the spectrum of *CBS* mutations resulting in homocystinuria.

Mutation	Position[a]	Exon	Nucleotide Change	Amino acid change	SIFT score	PolyPhen score	Prediction[b]	Mutation type[c]	Mutaion Status	Mutation presented in[d]
L136P	44486397	3	c.407T > C	L136P	0	1.0	Damaging	Het	Novel	III:2, III:3, III:4, III:6, II:4, II:6, I:4
A158V	44485784	4	c.473C > T	A158V	0	1.0	Damaging	Het	Novel	IV:1, III:2, III:3, III:4, III:5, II:3, I:1

Table 3. *CBS* mutations identified in this family with homocystinuria. [a]Genomic positions are presented according to NCBI build 36. [b]The SIFT and PolyPhenscore predict phenotypic effect. [c]Het: heterozygous mutation. [d]Subject number in this family with homocystinuria.

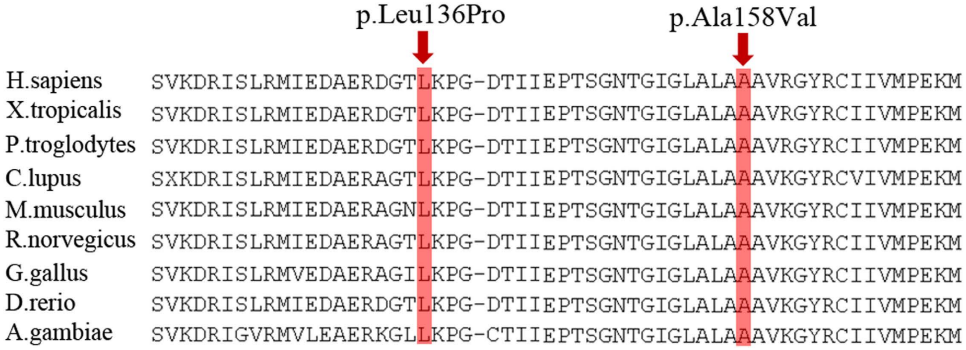

Figure 4. Orthologous protein sequence alignment of *CBS* from different species. The mutated residue showing conservation was shaded in red. Red shaded amino acids proteins showed that the two novel missense mutations occurred at highly conserved positions in these species.

The human *CBS* gene, located at chromosome 21q22.3[16], consists of 63-kDa subunits and encodes an enzyme with 551 amino acids[17]. The enzyme's structure consists of a catalytic domain with 409 amino acids in the N-terminal and a regulatory domain with 142 amino acids in the C-terminal[18]. The protein encoded by this gene acts as a homotetramer to catalyze the conversion of homocysteine to cystathionine, the first step in the trans-sulfuration pathway[1]. The encoded protein is allosterically activated by adenosyl-methionine and uses pyridoxal phosphate as a cofactor. Defects in this gene can cause *CBS* deficiency, which can lead to homocystinuria. And most of affected patients are compound heterozygotes of these *CBS* mutations[10,14,15,19]. Until now, molecular genetic analyses of *CBS* deficiency have identified more than 150 pathogenic mutations among homocystinuric patients, mostly in the Caucasian populations and very few in African-Americans and Asians[20]. In 2011, two *CBS* mutations (c.833T > C and c.1006C > T) were detected in a Hong Kong homocystinuric patient by Kwok *et al.*[19], however, it is so far the only report describing mutations in the *CBS* gene in Chinese (Fig. 2). In addition to the mutations identified in this study, the spectrum of mutations in *CBS* observed Han Chinese bears less resemblance to those observed in in Japanese and Korean patients[7,9].

In this study, mutation analysis of three patients with homocystinuria in a Han Chinese family is described and we identified novel compound heterozygous for mutations c.407T > C (p. L136P) in exon 3and c.473C > T (p.A158V) in exon 4 of the *CBS* gene (Fig. 3). So far, these two mutations are reported in homocystinuric patients for the first time in mainland Han Chinese, although *CBS* mutations have been identified in different ethnic groups. In this pedigree, all the three affected patients (III:2, III:3 and III:4) were found to harbor both of the two missense mutations in *CBS*. The patients in this family were diagnosed as homocystinuria based on detection of elevated blood homocysteine, and diagnosis of skeletal deformities, mental retardation and ectopialentis. The proband of this family (III:3) presented with reduced vision, and were diagnosed by Provincial Hospital Affiliated to Shandong University at the age of 15. Biocularlens dislocation, high myopia, exotropia, glaucoma and corneal staphyloma were proved at that time. She had a history of bilateral downward dislocation of the lens since 7years old. No significant family history was noted except her two sisters (III:2 and III:4), who exhibited similar clinical manifestations with the proband. Surgeries were performed to extract the dislocated lens during 8 to 11 years old in these three affected girls. Their plasma total homocysteine level and methionine level were both markedly elevated, confirming the diagnosis of homocystinuria (Table 3). Molecular genetic testing of the *CBS* gene also helps to confirm the diagnosis of patients. Mutation analysis was also performed on the patient's parents as well as her younger sister and brother, who are all unaffected (Fig. 1). Sequencing analysis showed that the father (II:3) and her sister (III:5)only carried thec.473C > T (p.A158V) mutation. Her mother (II:4) and brother(III:6) were heterozygous for thec.407T > C (p. L136P) mutation (Table 3).In addition, none of the two mutations in *CBS* was detected in 600 normal controls through gene analysis. Considering that *CBS* deficiency is an autosomal recessive disorder and that no other alteration was detected in coding regions of the *CBS* gene in homocystinuric patients of this family, it is highly possible that the two novel mutations of *CBS* identified here are responsible for the pathogenesis of homocystinuria in this pedigree.

For the p.L136P mutation identified in this pedigree, Leucine was replaced by Proline in exon 3, which is the most evolutionary conserved part of the *CBS* enzyme. Among all mutations identified in homocystinuric patients,

about 25% mutations were in this conserved region (the third exon of this gene)[5–12]. The p.A158V mutation of *CBS* resulted in a substitution of Alanine to Valinein exon 4. Moreover, they are both predicted to be probably damaging to protein function by SIFT and PolyPhen-2. However, the exact mechanisms and pathological roles of the two novel mutations in *CBS* in the development of homocystinuria are largely unknown. In addition, the spectrum of mutations observed in this study bears less resemblance to those observed in Japanese[9], Korean[7] and Filipino[10] patients, as well as Western countries, suggesting possible existence of ethnic differences among various populations. Future studies on Chinese homocystinuric subjects may help to provide more evidence for this hypothesis. In order to better understand homocystinuria pathogenesis, functional studies are needed to illustrate the role of *CBS* and the underlying mechanisms of this disease.

These data, together with the clinical presentation of the three affected siblings, demonstrated that p.L136Pandp. A158Vmutations in the *CBS* gene were responsible for homocystinuria in a Han Chinese family. Our data of *CBS* mutations causing homocystinuria further confirm the role of *CBS* in the pathogenesis of homocystinuria. This study expands the mutation spectrum of *CBS* resulting in homocystinuria, which could provide insights into the pre-symptomatic molecular diagnosis, the management of homocystinuric patients, and the genetic counseling of families in Chinese.

References

1. Miles, E. W. & Kraus, J. P. Cystathionine beta-synthase: structure, function, regulation, and location of homocystinuria-causing mutations. *J Biol Chem* **279,** 29871–29874 (2004).
2. Chamberlin, M. E. *et al.* Methionine adenosyltransferase I/III deficiency: novel mutations and clinical variations. *Am J Hum Genet* **66,** 347–355 (2000).
3. Mudd, S. H. *et al.* The natural history of homocystinuria due to cystathionine beta-synthase deficiency. *Am J Hum Genet* **37,** 1–31 (1985).
4. Kozich, V. & Kraus, J. P. Screening for mutations by expressing patient cDNA segments in E. coli: homocystinuria due to cystathionine beta-synthase deficiency. *Hum Mutat* **1,** 113–123 (1992).
5. de Franchis, R., Kraus, E., Kozich, V., Sebastio, G. & Kraus, J. P. Four novel mutations in the cystathionine beta-synthase gene: effect of a second linked mutation on the severity of the homocystinuric phenotype. *Hum Mutat* **13,** 453–457 (1999).
6. Gaustadnes, M. *et al.* The molecular basis of cystathionine beta-synthase deficiency in Australian patients: genotype-phenotype correlations and response to treatment. *Hum Mutat* **20,** 117–126 (2002).
7. Lee, S. J. *et al.* Identification and functional analysis of cystathionine beta-synthase gene mutations in patients with homocystinuria. *J Hum Genet* **50,** 648–654 (2005).
8. Urreizti, R. *et al.* Spectrum of *CBS* mutations in 16 homocystinuric patients from the Iberian Peninsula: high prevalence of T191M and absence of I278T or G307S. *Hum Mutat* **22,** 103 (2003).
9. Katsushima, F. *et al.* Expression study of mutant cystathionine beta-synthase found in Japanese patients with homocystinuria. *Mol Genet Metab* **87,** 323–328 (2006).
10. Silao, C. L., Fabella, T. D., Rama, K. I., Estrada, S. C. & Novel, C. B. S. gene mutations in a Filipino patient with Classical Homocystinuria. *Pediatr Int*, doi: 10.1111/ped.12666 (2015).
11. El-Said, M. F. *et al.* A common mutation in the *CBS* gene explains a high incidence of homocystinuria in the Qatari population. *Hum Mutat* **27,** 719 (2006).
12. Sebastio, G., Sperandeo, M. P., Panico, M., de Franchis, R., Kraus, J. P. & Andria, G. The molecular basis of homocystinuria due to cystathionine beta-synthase deficiency in Italian families, and report of four novel mutations. *Am J Hum Genet* **56,** 1324–1333 (1995).
13. Yang, Y. *et al.* Identification of a novel MYOC mutation in a Chinese family with primary open-angle glaucoma. *Gene* **571,** 188–93 (2015).
14. Martinez-Gutierrez, J. D., Mencia-Gutierrez, E., Gracia-Garcia-Miguel, T., Gutierrez-Diaz, E. & Lopez-Tizon, E. Classical familial homocystinuria in an adult presenting as an isolated lens subluxation. *Int Ophthalmol* **31,** 227–232 (2011).
15. Cozar, M. *et al.* Identification and functional analyses of *CBS* alleles in Spanish and Argentinian homocystinuric patients. *Hum Mutat* **32,** 835–842 (2011).
16. Munke, M., Kraus, J. P., Ohura, T. & Francke, U. The gene for cystathionine beta-synthase (*CBS*) maps to the subtelomeric region on human chromosome 21q and to proximal mouse chromosome 17. *Am J Hum Genet* **42,** 550–559 (1988).
17. Kraus, J. P. *et al.* The human cystathionine beta-synthase (*CBS*) gene: complete sequence, alternative splicing, and polymorphisms. *Genomics* **52,** 312–324 (1998).
18. Shan, X. & Kruger, W. D. Correction of disease-causing *CBS* mutations in yeast. *Nat Genet* **19,** 91–93 (1998).
19. Kwok, J. S. *et al. CBS* gene mutations found in a Chinese pyridoxine-responsive homocystinuria patient. *Pathology* **43,** 81–83 (2011).
20. Kruger, W. D., Wang, L., Jhee, K. H., Singh, R. H. & Elsas, L. J., 2nd. Cystathionine beta-synthase deficiency in Georgia (USA): correlation of clinical and biochemical phenotype with genotype. *Hum Mutat* **22,** 434–441 (2003).

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Author Contributions

Z.Y. and G.M. designed the study. C.Q., L.L., Z.L., Y.W., G.Z. and Y.X. recruited the participants. B.G., Y.S., X.L., F.H., X.F. and Z.Y. performed the genotyping. B.G. wrote the initial draft. Z.Y., Y.L. and X.L. corrected the English spelling and grammar. All authors critically revised, reviewed and gave final approval of this manuscript.

Additional Information

Competing financial interests: The authors declare no competing financial interests.

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