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BRIEF COMMUNICATION

Heterozygous Ala137Pro Mutation in Keratin 12 Gene Found in Japanese with Meesmann's Corneal Dystrophy

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Purpose: To report the molecular genetic analysis of a Japanese pedigree with Meesmann's corneal dystrophy (MCD).

Methods: Sequencing of the keratin 3 and keratin 12 genes was performed in 2 patients who were siblings and in an unaffected individual in the same family. The patients had the typical corneal microcysts and recurrent erosions with mild photophobia.

Results: A novel mutation resulting in the substitution of alanine to proline in codon 137 of the keratin 12 gene (Ala137Pro) was found in the 2 patients, but not in the unaffected member of the family and the 50 controls.

Conclusion: This novel mutation (Ala137Pro) of the keratin 12 gene found in a Japanese family had caused MCD. **Jpn J Ophthalmol 2002;46:673–674** © 2002 Japanese Ophthalmological Society

Key Words: Gene, keratin 12, Meesmann's corneal dystrophy, mutation.

Introduction

Meesmann's corneal dystrophy (MCD) is an autosomal dominant disorder characterized by a myriad of fine, round epithelial asymptomatic microcysts.[1,2] It is usually an asymptomatic disorder that is coincidentally detected in ophthalmic examinations. Occasionally there may be symptoms of corneal epithelial erosion. The keratin 3 gene (*KRT3*) and keratin 12 gene (*KRT12*) are candidate genes for the mutations that cause MCD because of their expression specificity. Irvine and co-workers identified heterozygous missense mutations in *KRT3* or *KRT12* in MCD families with European ancestors.[3] Nishida and his co-workers also found four mutations of *KRT12* in Japanese pedigrees.[4] Here we present a novel mutation of *KRT12* in a Japanese MCD pedigree.

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Materials and Methods

Cases

A 60-year-old man (II-5) had had photophobia since childhood. His visual acuity was 1.0 in both eyes without correction. He had a myriad of fine, round epithelial microcysts bilaterally (Figure 1). The epithelial surface was stained by fluorescein in both eyes. The corneal stroma was clear. Other ocular examinations revealed no abnormality. He had a 77-year-old sister (II-1) with similar ocular symptoms. Her visual acuity was 1.0 in the right eye and 0.6 in the left eye. There were multiple round microcysts in the corneal epithelium of both eyes. A cataract was detected in her left eye. Another sister (II-4) also had been diagnosed with MCD by a local ophthalmologist. The family pedigree is shown in Figure 2.

Genetic Analysis

This study was performed according to the tenets of the Declaration of Helsinki. After obtaining in-

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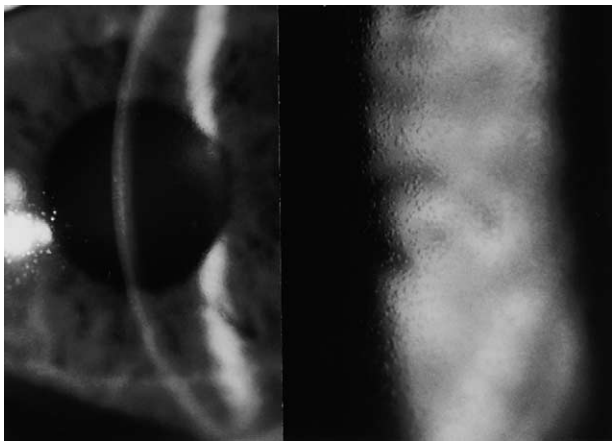

Figure 1. Slit-lamp micrographs show myriad fine, round corneal epithelial microcysts (left) in patient II-5 with Meesmann's corneal dystrophy. Higher magnification (right).

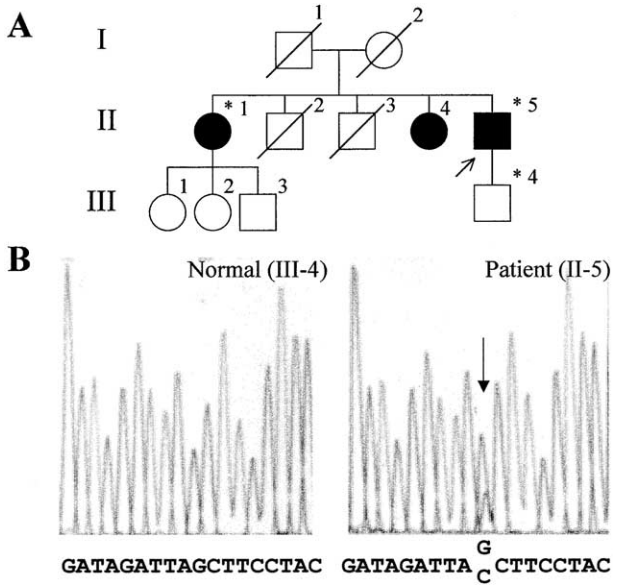

Figure 2. Mutation analysis of *KRT12* in the pedigree with Meesmann's corneal dystrophy (MCD). (**A**) Japanese pedigree with MCD. Blackened symbols represent affected individuals. Individuals from whom DNA was obtained are denoted by an asterisk (*). Arrow indicates the proband. (**B**) Sequence chromatograms of Exon 1 of *KRT12* from unaffected (left panel) and affected (right panel) family members. The right panel shows the heterozygous G to C transversion in codon 137, replacing alanine with proline (Ala 137 Pro) in patient II-5.

formed consent, genomic DNA was extracted from leucocytes that were collected from three members of the pedigree. Fifty persons without corneal disease also donated DNA after informed consent and served as controls. All exons of *KRT3* and *KRT12* were amplified from patient II-5 by using primers described elsewhere, with some modifications.[3,4] Amplified products were purified and sequenced using a cycle sequencing kit. Exon 1 of *KRT12* was also amplified from patient II-1, an unaffected member of the family III-4, and the 50 controls, using specific primers that were directly sequenced.

Results

The sequence analysis showed a (G/C) transversion- in the first nucleotide of codon 137 of *KRT12* in the 2 patients (Figure 2). The change would be expected to result in alanine (Ala) to proline (Pro) amino acid change (Ala137Pro). This alteration was not detected in the unaffected family member (III-4) and in the 50 unrelated controls. No mutation was detected in *KRT3* of the patients.

Discussion

We found a novel heterozygous Ala137Pro mutation of *KRT12* in a Japanese family with MCD. The mutation is located in the α-helix-initiation motif of the keratin protein. All known keratins have an alanine at the position corresponding to codon 137 of *KRT12*. Three different mutations at codon 135 in *KRT12*, which also are within the α-helix-initiation motif, have been reported.[3–5] Nishida and co-workers suggested that the mutations in the highly conserved keratin helix boundary motifs might cause a relatively severe form of MCD.[3] Ala137Pro change is also likely to cause symptomatic MCD phenotype with recurrent corneal erosion. Further genetic analysis of MCD patients will reveal additional genotype–phenotype relationships in keratin-related corneal disease.

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