



Hyaline Fibromatosis Syndrome: A Novel Mutation and Recurrent Founder Mutation in the *CMG2/ANTXR2* Gene

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Hyaline Fibromatosis Syndrome (HFS) is a rare autosomal recessive disorder affecting primarily skin and mucous membranes. Skin appears thickened with subcutaneous nodules, associated with swollen joint contractures, red hyperpigmentation and gingival hyperplasia. Additional findings include osteopenia and osteoporosis, and the affected children are susceptible to infections and protein losing enteropathy (1). Histopathology of skin lesions shows proliferation of spindle-shaped cells, embedded in a homogeneous hyaline-like material, and biochemical alterations in type I and VI collagens as well as in glycosaminoglycans have been reported (2, 3).

Initially, infantile systemic hyalinosis and juvenile systemic hyalinosis were considered as two distinct entities on the basis of time of onset. However, subsequent work demonstrated that both forms of hyalinosis were caused by mutations in the *CMG2* gene (also known as *ANTXR2*), indicating that these disorders are allelic and part of the same phenotypic spectrum, now known as HFS (4, 5).

The *CMG2* gene encodes a 55-kDa type I transmembrane protein known as capillary morphogenesis protein 2. While the precise physiologic function of this protein is currently unknown, its expression is upregulated in endothelial cells during capillary formation. This protein also serves as the main receptor of the anthrax toxin (6). The gene is expressed in all tissues with exception of the brain, a finding consistent with normal cognitive development of the affected individuals (6, 7).

CASE REPORTS

This study examines 4 cases with clinical features of HFS, all resulting from consanguineous marriages (Fig. 1 and Fig. S1¹). The clinical features of these patients are presented in Appendix S1¹. Following acquisition of informed consent in accordance with the Declaration of Helsinki Principles, DNA was extracted from peripheral blood samples taken from patients and family members (if available) using QIAamp DNA Blood Mini Kit (Qiagen, Valencia, CA, USA). PCR amplification of the *CMG2* gene was performed using 17 pairs of primers spanning all 17 exons and

the flanking intronic sequencing, followed by bidirectional Sanger sequencing. For haplotype analysis and homozygosity mapping, additional primers were designed for typing of 13 informed SNPs within and flanking the *CMG2* gene.

In Family 1, DNA was not available for mutation analysis from the deceased proband but both parents were heterozygous carriers of a previously unreported splice junction mutation (c.946-2A→G in intron 11) in *CMG2*, which by Human Splicing Finder program (www.umd.be/HSF/#) is predicted to result in aberrant splicing and a subsequent premature termination codon. Thus, the proband was deduced to be homozygous for this mutation. In Family 2, the proband had an insertion mutation, c.1073_1074insC (p.Pro358ProfsX13), which has been previously reported (8). This mutation in exon 13 causes a frameshift and results in premature termination of translation. In Family 3, a homozygous mutation, c.1074delT (p.Pro358ProfsX50) in exon 13, was detected; this recurrent mutation has been reported in a number of cases. In Family 4, no mutations in the *CMG2* gene were noted, and subsequent homozygosity mapping excluded this gene locus at chromosomal region 4q21 (Fig. S1¹).

Since the mutation c.1074delT has been encountered in a number of cases, we examined the possibility that this mutation is either a result of “founder effect” or is a “hotspot” mutation. For this purpose, haplotype analysis with 13 SNPs within and flanking the *CMG2* gene was performed with DNA from the proband in Family 2 as well as from another patient with HFS that we have recently described (9). The results revealed a 2 Mb conserved block within a 3 Mb region of the genome which included *CMG2* in these probands and suggesting a founder effect in these two Iranian cases of different ethnicity and language group.

DISCUSSION

Thus far, 37 distinct mutations have been identified in *CMG2* in ~150 HFS patients. This study adds a previously unreported mutation to this database. Furthermore, one of our patients demonstrated the same, recurrent mutation c.1074delT, which has previously been reported in a number of cases (9, 10). Haplotype analysis in the families reported here suggests that this mutation is a result of founder effect rather than being a hotspot mutation. Finally, extensive sequencing of the *CMG2* gene failed to reveal any pathogenic mutations in one family and homozygosity mapping excluded this gene. It should be noted that the latter patient had characteristic features of HFS, phenotypically similar to the other 4 cases, but also had additional features not present in other cases examined. These include immunodeficiency

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Fig. 1. Clinical features of patients with hyaline fibromatosis syndrome. Note a tumor on the right parietal area (a) and multiple nuchal papules (b) in Patient 3. Patient 4 had extensive perianal rash with subcutaneous nodules (c). Patient 2 had characteristic findings of HFS, including flexural contractures of the joints ("frog-leg position") (f), multiple reddish pearly papules on the face and neck (e, g), prominent low-set ears (e), and hyperpigmentation shown on lateral malleoli (d).

and infantile pyloric stenosis, features also present in the Jacobsen syndrome, known as 11q terminal deletion syndrome (11). Identification of mutations in the *CMG2* gene in families with HFS can be used to confirm clinical diagnosis and will form the basis for identification of heterozygous carriers in extended families with previous history of affected children. Finally, knowledge of the specific mutations allows prenatal testing and preimplantation genetic diagnosis to prevent the reoccurrence of HFS in consanguineous families with history of this disease.

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