Multiple Hereditary Exostoses: Report of an *EXT2* Gene Mutation in a Colombian Family

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J Pediatr Genet 2018;7:122–124.

Abstract

Keywords

- hereditary multiple exostoses
- EXT1
- EXT2
- osteochondroma
- children

Multiple hereditary exostoses (MHE) is a rare disease with autosomal dominant inheritance, caused by heterozygous germline mutations in the *EXT1* or *EXT2* genes. This disorder is characterized by the growth of prominences surrounded by cartilage in the growth plates and the long bones. Here, we report a family affected by MHE. In this family, a pathogenic variant c.544C > T (p. Arg182Ter) was identified in the *EXT2* gene. This variant has been previously described in the literature, and here we are reporting the relationship with clinical findings. MHE is suspected according to the clinical manifestations; molecular research should be performed to establish the most frequent mutations. A support, diagnosis, and follow-up group should be created, and genetic counseling should be available for patients and families.

Introduction

Multiple hereditary exostoses (MHE) is a disease characterized by growths of osteochondromas (OCs). At present, this disorder is called hereditary multiple OCs (HMO). Lesions may develop malignant characteristics and transform into chondrosarcoma, which can increase with age.¹ Here, we present a family with MHE; the molecular study was performed using direct sequencing of all encoding exons and multiplex ligation-dependent probe amplification (MLPA) of the *EXT1* and *EXT2* genes. No *EXT1* gene mutations were found, but the analysis of the *EXT2* gene showed evidence, in three individuals, of a heterozygous c.544C > T mutation that probably leads to the formation of nonfunctional *EXT2* proteins.

Case Report

We present the case of a Colombian family affected by MHE. All members of the family were born in Bogota, Colombia. Two siblings, the father and a paternal aunt had MHE. The

mother did not have any clinical manifestations of the disease, and there was no known history of the disease in the paternal grandparents (► Fig. 1). Case II4 is the 46-year-old father, who presented with OC growth at 13 years, showing progression until his 20s. Later, some of the tumors resolved spontaneously, and currently he has four in his lower and upper limbs, with skeletal deformities due to shortening of the right forearm. No other complications such as nerve, tendon, or vascular compression were noted.

Patient III2 is the first-born, 11-year-old male son. His tumor growth started at the age of 8 years in evident bony sites. At present, he has 10 OC localized mainly to the proximal fibula and tibia, distal femur (► Fig. 2), proximal humerus, and scapula. The lesions are hard, painless, elevated masses, 4 to 5 cm in size, and localized mainly to the epiphyses of the long bones. Case III3 is the youngest 8-year-old son. In his case, tumors began to appear when he was 2 years old. Currently, he has close to 14 painless OC localized to the proximal and distal femur and tibia, the proximal fibula and humerus, feet, and clavicles. None of the patients have developed malignant lesions.

received

November 5, 2017

accepted after revision

February 6, 2018

published online

March 7, 2018

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Verlag KG, Stuttgart · New York

DOI <https://doi.org/10.1055/s-0038-1636998>.
ISSN 2146-4596.

Fig. 1 Pedigree analysis for MHE (# age of the case report, *age of onset of symptoms). MHE, multiple hereditary exostoses.

According to the phenotypic characteristics described in the family, we suggest autosomal dominant MHE. The molecular study was performed using direct sequencing of all encoding exons and MLPA of the *EXT1* and *EXT2* genes from peripheral blood of all members of the family. The analysis did not reveal *EXT1* mutations, but direct resequencing of the *EXT2* gene showed that the father and the two siblings had a cytosine–thymine substitution in position 544 of exon 3 (c.544C > T) of heterozygous form. This mutation creates a premature stop codon (p.Arg182Ter) that leads to the formation of nonfunctional *EXT2* proteins.^{1–3} This is a hotspot mutation that is found in 4% of patients with *EXT2* muta-

tions.⁴ This mutation affects the far N-terminus of *EXT2*, and the produced truncated protein lacks the entire conserved C-terminal domain of *EXT2*. The mutation results in an unstable mRNA, and OC formation may be due to the loss of function produced by the truncated protein or haploinsufficiency caused by nonsense-mediated mRNA decay.⁴

The molecular analysis confirms the diagnosis in this Colombian family, and all family members were given genetic counseling.

Discussion

MHE is a disease with autosomal dominant inheritance and its exact prevalence is unknown considering that some cases are asymptomatic or subclinical, but it is estimated at around 1 in 50,000 to 100,000 in the Caucasian population.^{1–6} The prevalence in Colombia is unknown. The ratio between females/males is 1.5:1.^{3,7} OCs are the most frequent benign tumors of the bones, and nearly 80% manifest in the first 10 years of life due to chondrocyte proliferation and bone growth in the metaphyseal region in children.^{8,9}

MHE is caused by pathogenic variants in the *EXT* gene family that encode for exostosin proteins (EXT). The *EXT1* gene consists of 11 exons, the *EXT2* gene of 16 exons, and *EXT3* gene is unknown. These genes are located on chromosome 8q24.11–q24.13, 11p12–p11, and 19p, respectively.^{3,8} These proteins are glycosyltransferases that form a hetero-oligomeric complex that catalyzes elongation of the heparan sulfate chains.^{2,5,6}

The *EXT1* gene mutations range between 44 and 70%, and the *EXT2* gene mutations between 27 and 40% of cases.^{2,8} Inactivation mutations are the most common and account for 80% of cases and, by order of frequency, they include frame shift, nonsense, splice-site, and missense mutations.^{5,8} OC arise from bones with endochondral ossification; they may

Fig. 2 OC in the right lower limb. Rx of the femur: osteochondroma of the femoral neck and the fibula. OC, osteochondroma.

change in number and size and be bilateral or unilateral and symmetrical. They grow during the first decade of life and locate to the metaphyses of long bones.^{2,5,8} In general, patients may be asymptomatic, or present with limb angulations or short stature. Severe deformities can occur in up to 40 to 60% of patients.⁹ Clinical manifestations depend on the deformity and involvement of the neighboring structures.

Spontaneous regression of a solitary OC has been described. The majority of these lesions were reported in young males, as in case II4 of this report.¹⁰ In less than 5% of cases there will be malignant transformation to chondrosarcoma. For this reason, patients must be followed closely, especially in cases of OC that appear after puberty.¹¹

Diagnostic criteria include at least two OC in the juxta-epiphyseal region of the long bones with a family history of MHE and/or a mutation in one of the *EXT* genes.^{5,7} Molecular diagnosis must be made of the *EXT1* or *EXT2* gene mutation to confirm the mutation and to assess the degree of phenotype/genotype relationship. Differential diagnoses to be ruled out include Ollier's disease, chondrosarcomas, multiple epiphyseal dysplasia, and periosteal chondroma.⁴ The goal of treatment is to manage symptoms, and asymptomatic lesions require close follow-up and surveillance. Lesions requiring surgical management must be individualized to symptomatic patients with MHE and OC.^{2,5,6,8}

In the family described here, the paternal grandparents were not affected. However, as two of their offspring had OCs, this may have occurred from germinal mosaicism in one of the paternal grandparents (I1, I2). Nevertheless, it was not possible to obtain DNA samples from any of these patients (I1, I2, II2) to corroborate this. On the other hand, in MHE the penetrance is approximately 96% in females and 100% in males, and in 10% of affected individuals the result is from a *de novo* pathogenic variant.^{8,12} In this family, it was not possible to rule out that the paternal grandmother had a less severe clinical phenotype or reduced penetrance.

Likewise, the number of OCs that develop in an affected person varies widely even within families, and the involvement is usually symmetric,^{13,14} which could explain why the children have more severe clinical characteristics than the affected father. The somatic study of the tumors could be of help to obtain insights about the molecular biology of the tumor, which allows evaluation for the presence of other somatic mutations involving alternative signaling pathways that worsen the phenotype in the offspring.

To conclude, our Colombian family had a nonsense-type heterozygous mutation in exon 3 of the *EXT2* gene (c.544C > T), which generates an unstable mRNA, premature protein termination, or truncated protein, leading to OC formation.¹² This mutation was previously reported by Dobson-Stone et al in 2000, in patients with MHE in the United Kingdom.³ However, in Colombia, this is the first report of patients with MHE and mutations in exon 3 of the *EXT2* gene.

There are few clinical reports of MHE in Colombia and the incidence of the disease has not been described, hence the

need to determine which are the most frequent mutations in the Colombian population. This type of case report can help with the description of the molecular epidemiology of multiple exostoses in our country, to aid with genetic counseling.

Authors' Contributions

All authors were involved in drafting and translation of the manuscript.

Acknowledgments

Special thanks to Fundación Universitaria de Ciencias de la Salud, Hospital de San José, Bogotá D.C. Colombia. We also thank Dr. Wim Wuyts (Department of Medical Genetics–University and University Hospital of Antwerp, Belgium) for processing patient samples.

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Erratum: The article has been corrected as per Erratum (DOI: 10.1055/s-0042-1757504).