

CORRESPONDENCE



GGPS1 Mutation and Atypical Femoral Fractures with Bisphosphonates

TO THE EDITOR: Atypical femoral fractures have been associated with long-term bisphosphonate treatment.^{1,2} However, the underlying mechanisms remain obscure. We studied three sisters who had atypical femoral fractures after receiving various oral bisphosphonates for 6 years. Two of the sisters had a single fracture (at the ages of 64 and 73 years), and one had bilateral fractures (one at the age of 60 years and the other at the age of 61 years). Given the low incidence of atypical femoral fractures in the general population (5.9 per 10,000 person-years),³ we hypothesized that these sisters might have an underlying genetic background that contributed to these fractures.

We performed whole-exome sequencing to detect possible shared genetic variants involved in their apparent increased risk. In addition, we performed whole-exome sequencing in three unrelated patients with atypical femoral fractures who each had received bisphosphonates for more than 5 years. We prioritized rare nonsynonymous mutations in the variant filtering, and only mutations that were shared among the three sisters were considered. No mutation was found to be

homozygous or in any gene containing mutations in both chromosomes (compound heterozygous). Assuming that a dominant model was involved, we detected 37 rare mutations (in 34 genes), among them a novel p.Asp188Tyr substitution in the enzyme geranylgeranyl pyrophosphate synthase (GGPPS), which is a site of inhibition by bisphosphonates in the mevalonate pathway.⁴ The variant that is located in the genomic position g.235505746G→T on chromosome 1 (GRCh37/hg19) in *GGPS1* had the best conservation score and was not described in any of the available databases. This variant would be expected to severely impair the enzyme activity (Fig. 1). Furthermore, the gene encoding cytochrome P-450 family 1 subfamily A member 1 (*CYP11A1*), which is involved in steroid metabolism, was also mutated in all three sisters and in one of the unrelated patients, which suggests that it could be another potential susceptibility gene for bisphosphonate-related atypical femoral fractures. An additional mutation in the gene encoding mevalonate diphosphate decarboxylase (*MVD*) was detected in one unrelated patient.

Pathway analysis of the mutated genes showed enrichment of the isoprenoid biosynthetic pathway (GO:0008299), which includes *GGPS1*, *CYP11A1*, and *MVD* ($P < 0.001$). We speculate that other variants that have been identified might also be involved in susceptibility to bisphosphonate-related atypical femoral fractures. Such variants include missense changes in the gene encoding fibronectin 1 (*FNI*) and in the genes encoding synapse defective Rho GTPase homolog 2 (*SYDE2*) and neuronal guanine nucleotide exchange factor (*NGEF*); the latter two proteins are regulators of small GTPases. We speculate that our results may support a model in which accumulation of susceptibility variants (including some in rele-

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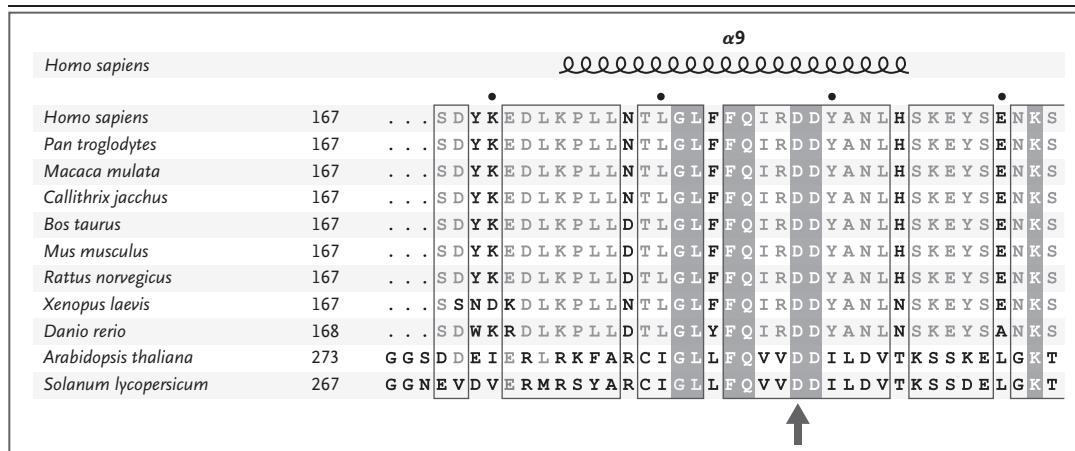


Figure 1. High Degree of Conservation of the Relevant Amino Acid Sequence of GGPS1 in Multiple Species.

Shown is the alignment of the region containing the D188 residue (arrow) in geranylgeranyl pyrophosphate synthase protein encoded by *GGPS1* in different species. This mutation would be expected to severely impair enzyme activity by disrupting a Mg^{2+} binding site that is critical for binding of the farnesyl pyrophosphate substrate and for catalysis.⁵ This mutation would be expected to disrupt the α -helix secondary structure (shown above the corresponding sequences). The numbers after each species indicate the first identified residue of the corresponding protein.

vant genes, notably *GGPS1*) may lead to a possible genetic component of predisposition to atypical femoral fractures.

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