A recurrent missense mutation of GDF5 causes proximal symphalangism in an British family.

Response to reviewers

Reviewer A

Comments

1. I would prefer to name the mutation in the title, like: Proximal symphalangism caused by a missense mutation in GDF5 (p.R438L) in a British family

2. SYMB1 should be SYM1B throughout the manuscript

3. Abstract: Skip SYM1B in the abstract, and just write about proximal symphalangism in general, because you don't know in the beginning if it is SYM1B caused by GDF5 mutations or SYM1A caused by NOG mutations.

4. "This report highlights the importance of thorough history taking, including a three generation family history, and detailed clinical examination of children with fixed planovalgus feet and other family members to detect rare skeletal dysplasia conditions causing pain and deformity, and provides details of the spectrum of problems associated with SYMB1." This argument is not conclusive and should be rephrased.

5. Introduction: "…NOG, encoding noggin, and GDF5, encoding growth differentiation factor 5" should be "NOG, encoding NOG, and GDF5, encoding Growth Differentiation Factor 5"

6. Case Report: The first sentence is identical with a sentence in the abstract and should be rephrased.

7. "DNA sequencing of the eldest son identified a heterozygous missense mutation c.1313G>T in GDF5, which causes the amino acid substitution arginine to leucine at codon 438 (R438L) which predicted to disrupt GDF5 function." R43L does not disrupt GDF5 function. R438L alters GDF5 signalling.
8. NOGGIN should be abbreviated as NOG throughout the manuscript.

9. Genes and proteins does not follow the standard nomenclature http://www.genenames.org/about/guidelines

10. "transforming growth factor superfamily" should be "transforming growth factor beta superfamily"

11. "The c.1313G>T (R438L) mutations in GDF5 has been previously detected in a family with SYM1 (5) and the causative mechanism is thought to be overexpression of GDF5 leading to joint cartilage overgrowth and subsequent fusion.(1, 4, 5)" R438L has been previously identified in a family with SYM1 and SYNS. The causative mechanism is not due to overexpression, but due to increased biological activity.

12. Table 2: Dawnson et al should be Dawson et al

13. Figures: more radiographs, especially from the hands would be helpful to evaluate the phenotypic variations in more detail.

Response to Reviewer

1. Added as requested
2. Amended as requested
3. We thank the reviewer for their comment. We prefer to keep the current structure as this case report focuses on the SYM1B mutation.
4. We thank the reviewer for their comment. We believe that this case report highlights the importance of family history and thorough systemic clinical examination.
5. Changed in introduction as requested.
6. Text altered at case report as requested.
7. Texted amended at Case report to include the effect of R438L on signalling.
8. Noggin abbreviated as NOG throughout the manuscript as requested
9. We thank the reviewer for their comment. Two of the authors of the manuscript are geneticists. We believe we have followed standard nomenclature but we are open to suggestions in the case we have deviated.
10. Text added in page No 6 as requested
11. Text amended in page No 6 as requested
12. Dawson spelling changed in Table 2 as requested
13. We thank the reviewer for their comment. We have attached all the available hand radiographs.

Reviewer B

Comments

1. Please have a figure showing the sequencing electropherogram of the mother and son, together with an homozygous control (potentially the father). A multi-species alignment of this mutated region showing the conservation of the R438 would also also add to this figure .

2. If you have DNA from the other affected children, please show evidence that they have the same mutation.

3. Please provide additional data on the predicted effect of the mutation on GDF5 function e.g. PolyPhen/Mutation taster prediction, details of the protein domain where the mutation is located and

Response

1. We thank the reviewer for their comment. The sequencing electropherogram is not readily available regretfully.

2. We thank the reviewer for their comment. We do not hold any further DNA other than the one stated in the manuscript.

3. The following text has been added in page 6.

The p.Arg438Leu substitution is a gain-of-function mutation known to be associated with proximal symphalangism (Seemann et al., 2005). Compared to wild type GDF5, the resulting protein shows increased biological activity that alters receptor-binding affinity within the TGFB signalling pathway. Overexpression of GDF5 disrupts normal joint formation and causes proximal symphalangism.