

LETTER TO THE EDITOR

A 24-generation-old founder mutation impairs splicing of *RBBP8* in Pakistani families affected with Jawad syndrome

Dear Editor,

Jawad syndrome (JWDS) (MIM #251255) is a multiple congenital anomaly and intellectual disability (ID) syndrome characterized by congenital microcephaly, facial dysmorphism, digital anomalies, onychodysplasia, and white spots on hands and feet present in all adult patients. So far, only two families have been reported, hereafter referred to as F1 and F2.^{1,2} Both families originate from Pakistan and carry mutation (NM_002894.3:c.1808-1809delTA;p.[Ile603Lysfs*7]) in *RBBP8*.²

We recruited two new families (F3 and F4) afflicted with JWDS from distant areas of Pakistan. After getting informed consents from parent(s)/guardians, clinical details were documented. Approval of this study was obtained by the Ethical Review Board of the National Institute for Biotechnology and Genetic Engineering (NIBGE), Pakistan. In F3, phenotypic variability with respect to height, digital anomalies, white spots, and nail dysplasia was observed (Figure 1(A)). F4 showed variable expression of onychodysplasia and white spots (Figure 1(A)).

Exome sequencing of a patient from F3 revealed above-mentioned *RBBP8* variant. Using Sanger sequencing, we found it also in F4 (Figure 1(B)).

To analyze mutational effects on splicing, we amplified mutation spanning region, which showed an expected band size of ~390 bps in all three samples. However, the patients showed an additional band of ~260 bps (Figure 1(C)). Sequencing of ~390 bps amplicons showed a frameshift and a premature termination codon (p.[Ile603Lysfs*7]) whereas ~260 bps revealed an in-frame deletion as the consequence of the skipping of exon 12, which entails a loss of 43 amino acids, p.(Ile603_Gln646delinsLys) (Figure 1(D,E)).

To assess the possibility of a founder mutation, all four families were genotyped. Haplotype analyses revealed a block of concordant SNP alleles of 1.06 Mb in the region of *RBBP8* that turned out to be shared by all four Pakistani JWDS families (Figure 1(F)). Based on a calculation described earlier,³ we estimate the minimum age of the mutation to be 24 generations, which corresponds to about 600 years assuming a generation time of 25 years (Figure 1(F)).

The identified mutation in *RBBP8* is, obviously, loss-of-function; however, depletion of *Rbbp8* in mice cause embryonic lethality.⁴ We propose that the transcript with the 2-bp deletion is degraded by nonsense-mediated decay (Figure 1(G)) which may encode *RBBP8* with residual function. This seems to explain the alleviated phenotype, since the missing amino acids are far from functionally important domains (Figure 1(E)).

Our findings add important details to the clinical spectrum of this mutation and highlight it as a founder mutation.

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CONFLICT OF INTEREST



The authors declare no conflict of interest.

PEER REVIEW

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DATA AVAILABILITY STATEMENT

Available on request to corresponding author.

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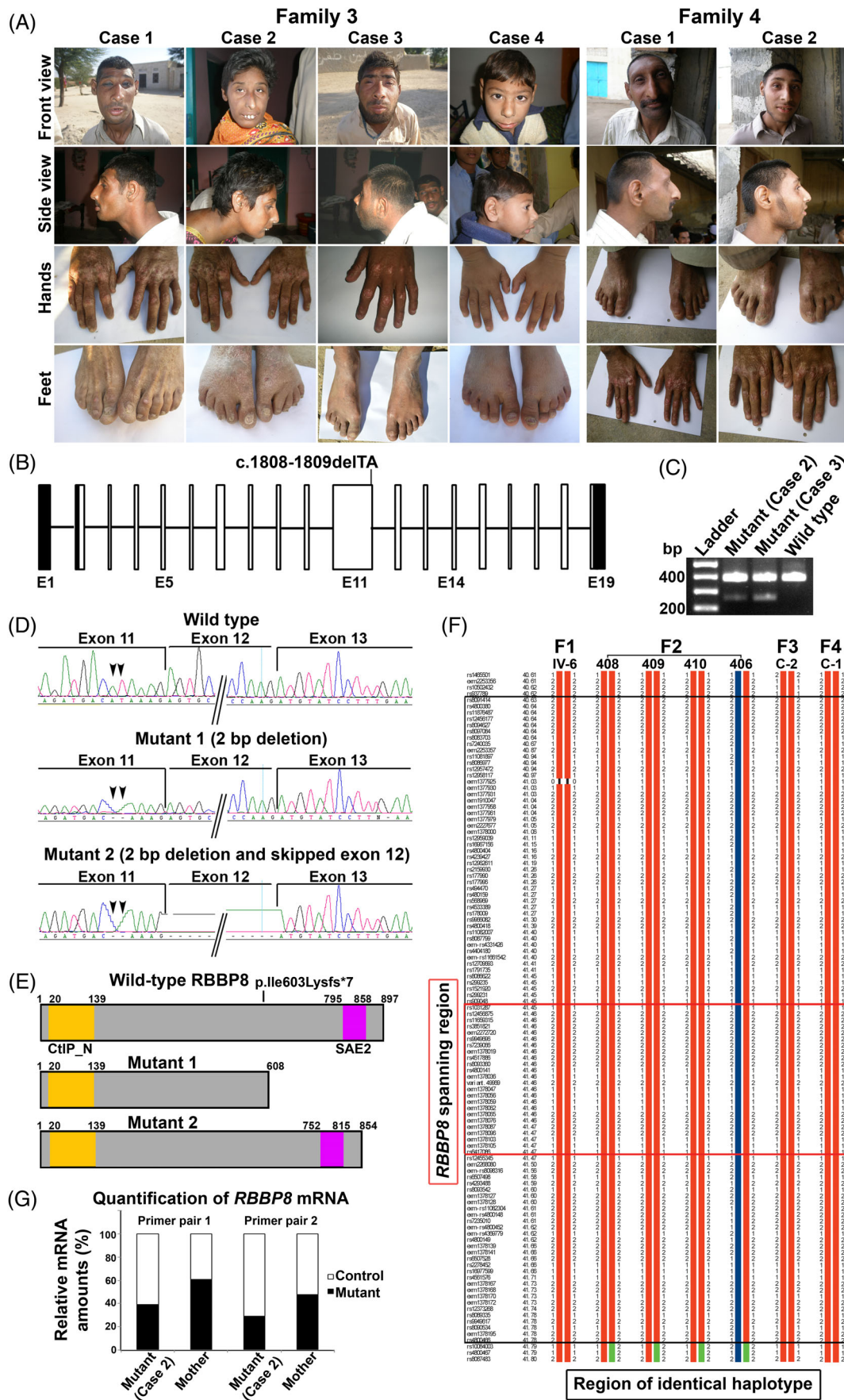


FIGURE 1 (A) Clinical features of F3 and F4. (B) Graphical representation of *RBBP8*. (C) Electropherogram showing transcript amplicons obtained from cDNA of two patients and a control. (D) Sanger traces corresponding to transcript amplicons of 387 bps (Mutant 1) and 260 bps (Mutant 2) from a patient in comparison to control. (E) Graphical illustration and domain structure of the RBBP8 protein. (F) Shared haplotype of four patients from four different families. (G) Quantitative real-time PCR of *RBBP8* mRNA [Colour figure can be viewed at wileyonlinelibrary.com]

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