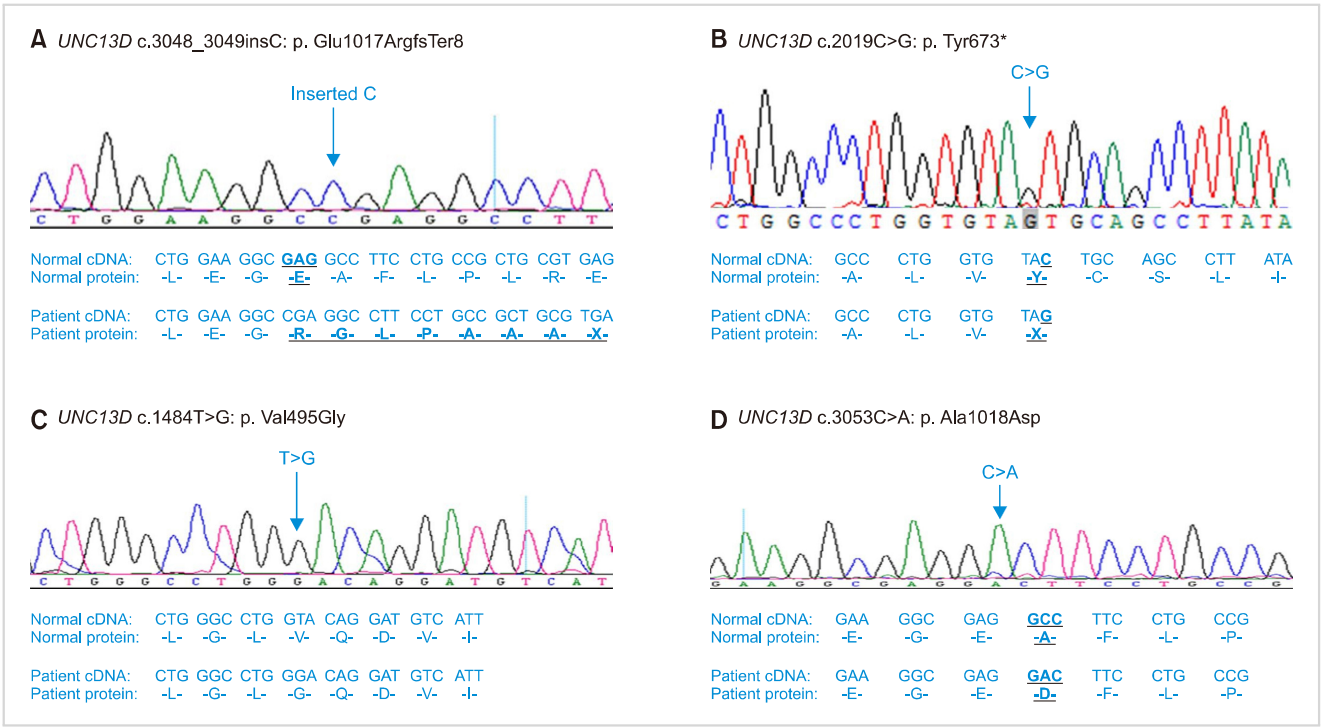
Supplement 1. Additional genetic methods and materials

Nucleotide sequences of all exons and flanking intronic sequences of *PRF1, UNC13D, STX11, STXBP2, LYST, RAB27A, SH2D1A*, and *XIAP* were amplified by PCR, followed by direct sequencing. PCR was performed using 30 ng of genomic DNA, 0.3 μmol each of the forward and reverse primers, 0.2 mmol each dNTP, 1.5 mmol/L MgCl2, 2 Units of Taq DNA Polymerase, and 2× PCR buffer in a total volume of 25 μL. Touchdown PCR was used to amplify all of the exons in the same reaction plate. Reaction conditions were as follows: initial denaturation for 15 min at 95°C, followed by multiple cycling in which the annealing temperature was gradually reduced. Each cycle consisted of 5 cycles of 95°C for 45 s, annealing at 67°C for 45 s and reduced by 2°C every cycle, and 72°C for 1 min. This was followed by an additional 29 cycles of denaturation at 95°C for 45 s, annealing at 45°C for 45 s, and elongation at 72°C for 1 min, and a final extension for 10 min at 72°C. Primer sequences are available upon request.

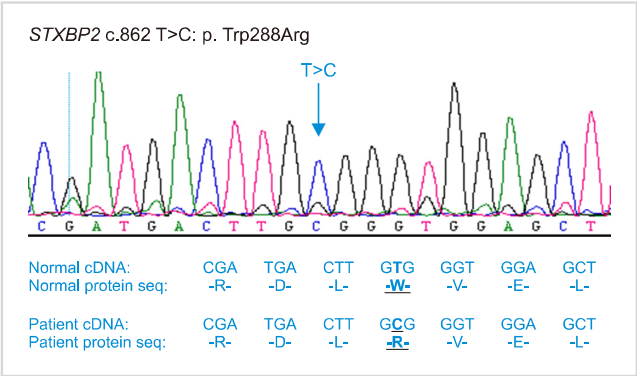
A mutation analysis was performed by direct sequencing of PCR fragments obtained by touchdown PCR using ABI 3730 Genetic Analyzers (Applied Biosystems, Foster City, CA, USA). The data were analyzed using DNA SeqMan (Lasergene 6). Sequence variants, if any, were analyzed with reference to the wild-type sequences (GenBank accession no. NM_199242 for *UNC13D*, NM_003764 for *STX11*, NM_001272034 for *STXBP2*, NM_000081.3 for *LYST*, and NM_004580 for *RAB27A*).

Supplementary Table 1. Pathogenic assessment of seven missense novel variants identified in our cohort.

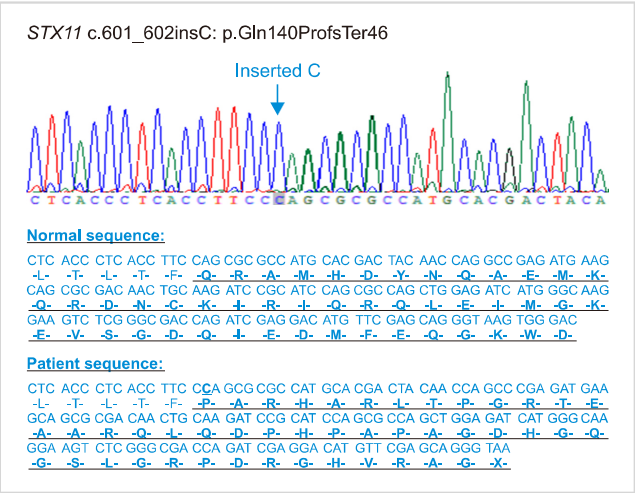
Genes	Novel variants	Saudi controls	MutationTaster	PolyPhen-2	SIFT	CADD
UNC13D	c.1484T>G: p. (Val495Gly)	0	Disease causing	Probably damaging	Damaging/100%	25.1
	c.3053C>A: p. (Ala1018Asp)	0	Disease causing	Probably damaging	Damaging/100%	28.4
STXBP2	c.862T>C: p. (Trp288Arg)	0	Disease causing	Probably damaging	Damaging/100%	24.5
	c.1034C>T: p. (Thr345Met)	0	Disease causing	Probably damaging	Tolerated: 100%	25.2
STX11	c.690G>A: p. (Gln230Ala)	0	Disease causing	Probably damaging	Damaging/100%	11.81
LYST	c.4637C>T: p. (Ala1546Val)	0	Disease causing	Probably damaging	Damaging/100%	22.8
RAB27A	c .400A>C: p. (Lys134Gln)	0	Disease causing	Probably damaging	Damaging/100%	27.7



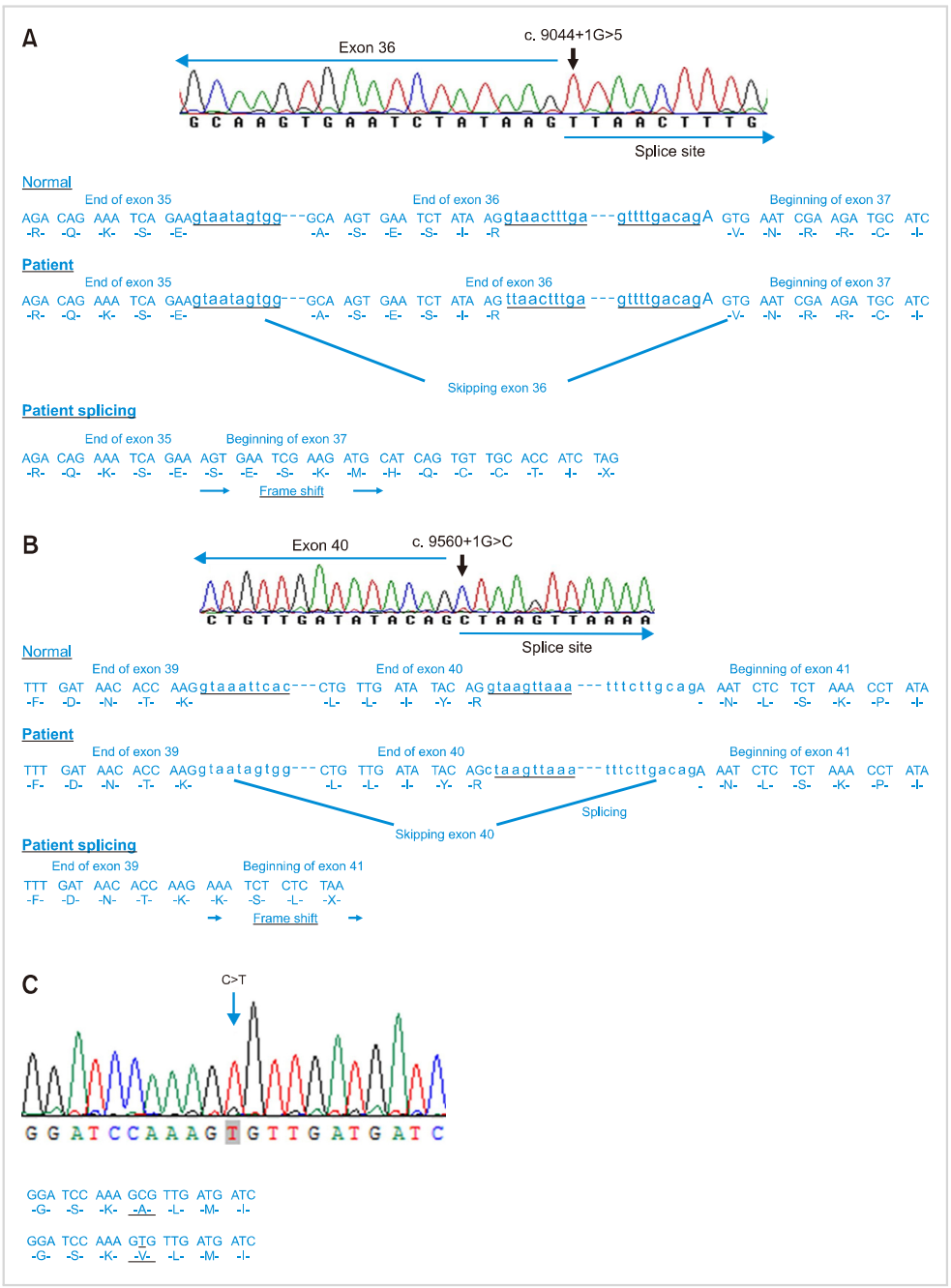
Supplementary Fig. 1. **(A)** *UNC13D* c.3048_3049insC: p. Glu1017ArgfsTer8, **(B)** *UNC13D* c. 2019C>G: p. Tyr673*, **(C)** *UNC13D* c.1484T>G: p.Val495Gly, **(D)** *UNC13D* c.3053C>A: p. Ala1018Asp.



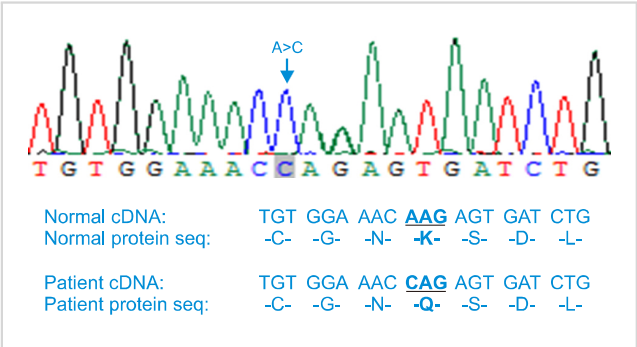
Supplementary Fig. 2. *STXBP2* c.862 T>C: p. Trp288Arg.



Supplementary Fig. 3. *STX11* c.601_602insC: p.Gln140ProfsTer46.



Supplementary Fig. 4. **(A)** *LYST* c.9044+1 G>T, **(B)** *LYST* c.9560+1 G>C, **(C)** *LYST* c.4637 C>T: p.Ala1546Val.



Supplementary Fig. 5. *RAB27A* c.400A>C: p. Lys134Gln.