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A novel missense variant in the *PNPLA1* gene underlies congenital ichthyosis in three consanguineous families

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Editor

Autosomal recessive congenital ichthyosis (ARCI) encompasses several forms of non-syndromic ichthyosis characterized by a rare, heterogeneous group of skin disorders affecting cornification.[1] To date, nine genes responsible for causing ARCI have been identified.[2–4]

In this study, we have recruited three unrelated consanguineous Pakistani families presenting features of ARCI for genetic analysis. The study was carried out according to the protocol approved by the Institutional Review Boards of the Quaid-i-Azam University, Islamabad, Pakistan and Baylor College of Medicine and Affiliated Hospitals, Houston, Texas, USA. All participants or legal guardians provided written informed consent.

Affected members in all the three families shared common characteristic features of generalized, fine, dry, dark brown scaling of the body with abnormal sweating and hyperthermia. Affected members in the family manifested mild erythroderma of skin lesion (Fig. 1d,e), in family b exhibited mildly hyperkeratotic skin around knees with exfoliation of entire skin (Fig. 1f,g) and in family c showed dark brown scaling severely affecting neck and face with exfoliative dermatitis (Fig. 1h,i). Conditions such as Ectropion, eclabium and alopecia were not observed in affected members of the families.

In two families (a and b), genome-wide SNP-based genotyping and exome sequencing were carried out as described previously.[5] In family c, microsatellite markers-based genotyping and dideoxy chain termination sequencing were performed as described by Mehmood *et al*.[6]

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Analysis of SNPs genotyping in two families (a and b) and microsatellite markers generated haplotype in family c established a linkage on chromosome 6p22.3-p21.2. Exome and Sanger dideoxy chain termination sequencing revealed a novel transversion variant c. 102C>A (p.Asp34Glu) in the *PNPLA1* gene located on chromosome 6p21.31 that co-segregated with ARCI in the families. Additionally, the *PNPLA1* c.102C>A (p.Asp34Glu) variant was found absent in the Exome Aggregation Consortium database, in 94 unrelated Pakistani control samples and in 215 in-house exome sequences from unrelated Pakistani individuals without ichthyosis. The *PNPLA1* c.102C>A (p.Asp34Glu) variant occurs at a residue that is highly conserved from human to *C. elegans* (Fig. 2i). The pathogenic nature of the missense variant p.Asp34Glu was also supported by protein prediction tools PolyPhen-2.1 (http://genetics.bwh.harvard.edu/pph/) and SIFT (http://sift.jcvi.org) envisaging the variant is probably damaging (Table 1).

In the present study, we have provided the first evidence of involvement of *PNPLA1* gene in three unrelated consanguineous Pakistani families segregating ARCI. To date, only three pathogenic sequence variants have been reported in the *PNPLA1* gene (Table 1). Here, we have reported the fourth novel and third missense variant (c.102C>A, p.Asp34Glu) in the *PNPLA1* gene in three unrelated families of Pakistani origin.

Patatin-like phospholipase domain containing (PNPLA) is a mammalian protein family consisting of nine members, including PNPLA1, and involved in lipid metabolism and signal pathway, and exhibits diverse lipolytic and acyltransferase activities.[8] PNPLA1 is present in the epidermis and strongly expresses in the granular layer which is associated with accumulation of lipid droplets in the keratinocytes. Sequence variants in the *PNPLA1* gene abolish lipid droplet accumulation in the keratinocytes suggesting PNPLA1 function not in triglyceride hydrolase activity but in glycerophospholipid synthesis or remodelling, which could be mediated by acyl-CoA-dependent or independent acyltransferase activity.[3] All the four mutations including three reported previously[3,7] and the one (p.Asp34Glu) identified in the present study (Table 1) affect the same conserved patatin domain which features the catalytic dyad (Asp 172 and Ser53) and cause ARCI.[8]

The 3D models of normal and mutated PNPLA1 were predicted using homology modelling (SWISS-MODEL software[9]) and characterized by online structure analysis tools (Swiss-PdbViewer). The primary protein sequence of human PNPLA1 was obtained from the UniProt database (www.uniprot.org). The protein modelling studies indicated that p.Asp34Glu variant occurs within the patatin domain of the PNPLA1 protein. The negatively charged glutamate has an extra side chain and extra rotatable hydrogen bond, a heavy atom counter and an additional undefined bond stereocentre. Thus, the alteration (p.Asp34Glu) might result in distorting the structure of PNPLA1 protein due to differences in the structures of the involved amino acids (Fig. 2f–h).

In conclusion, the variant identified will expand the spectrum of mutations in the *PNPLA1* gene, provides more evidence for lack of genotype–phenotype correlation and clinical variability in *PNPLA1* and underscores its role in causing ARCI.

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References

1. Oji V, Tadini G, Akiyama M, et al. Revised nomenclature and classification of inherited ichthyoses: results of the First Ichthyosis Consensus Conference in Sorèze 2009. J Am Acad Dermatol. 2010; 63:607–641. [PubMed: 20643494]
2. Fischer J. Autosomal recessive congenital ichthyosis. J Invest Dermatol. 2009; 129:1319–1321. [PubMed: 19434086]
3. Grall A, Guaguère E, Planchais S, et al. PNPLA1 mutations cause autosomal recessive congenital ichthyosis in golden retriever dogs and humans. Nat Genet. 2012; 44:140–147. [PubMed: 22246504]
4. Radner FP, Marrakchi S, Kirchmeier P, et al. Mutations in CERS3 cause autosomal recessive congenital ichthyosis in humans. PLoS Genet. 2013; 9:e1003536. [PubMed: 23754960]
5. Ansar M, Raza SI, Lee K, et al. A homozygous missense variant in type I keratin KRT25 causes autosomal recessive woolly hair. J Med Genet. 2015; 52:676–680. [PubMed: 26160856]
6. Mehmood S, Jan A, Muhammad D, et al. Mutations in the lipase-H gene causing autosomal recessive hypotrichosis and woolly hair. Australas J Dermatol. 2015; 56:66–70.
7. Fachal L, Rodríguez-Pazos L, Ginarte M, Carracedo A, Toribio J, Vega A. Identification of a novel PNPLA1 mutation in a Spanish family with autosomal recessive congenital ichthyosis. Br J Dermatol. 2014; 170:980–982. [PubMed: 24344921]
8. Kienesberger PC, Oberer M, Lass A, Zechner R. Mammalian patatin domain containing proteins: a family with diverse lipolytic activities involved in multiple biological functions. J Lipid Res. 2009; 50:63–68.
9. Schwede T, Kopp J, Guex N, Peitsch MC. SWISS-MODEL: an automated protein homology-modeling server. Nucleic Acids Res. 2003; 31:3381–3385. [PubMed: 12824332]

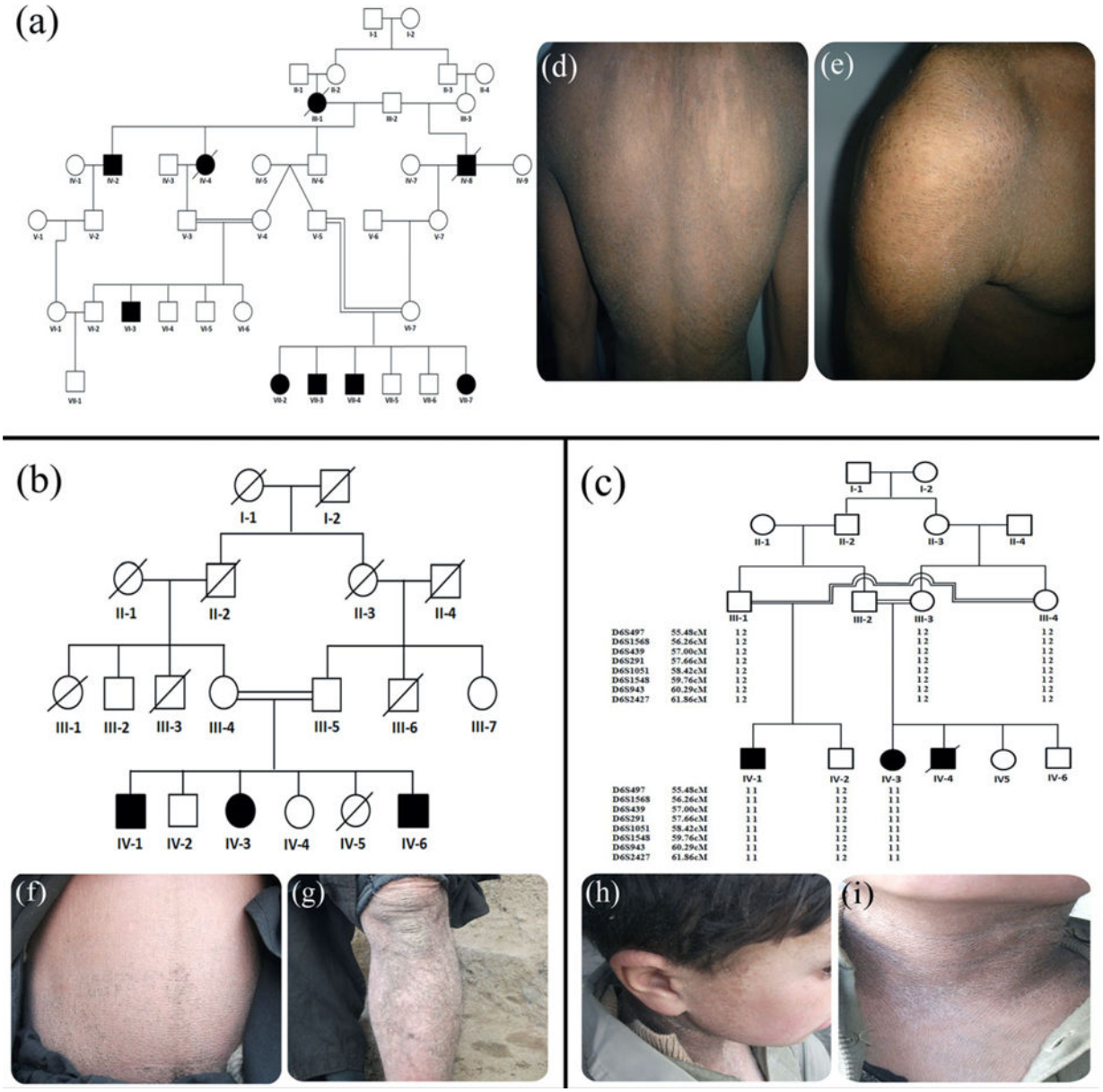

Figure 1.

Pedigrees of the three consanguineous families segregating autosomal recessive congenital ichthyosis (ARCI). Double lines indicate a consanguineous union. Clear symbols represent unaffected and filled symbols affected individuals. A cross line on the symbol indicates a deceased individual. Clinical features of affected members are presented in panels d and e (family a), f and g (family b), h and i (family c).

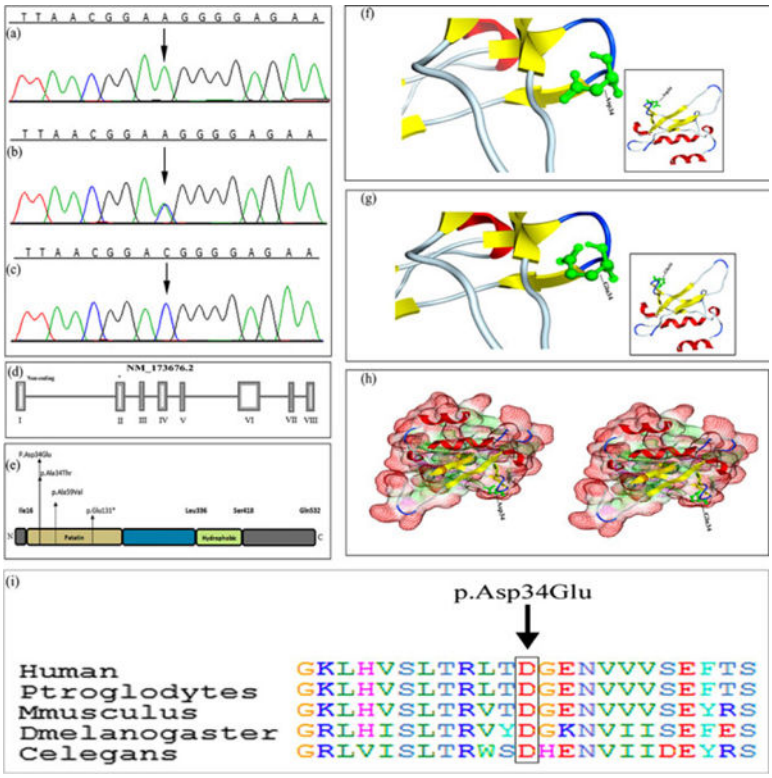

Figure 2.

Characteristics of the novel missense variant (c.102C>A, pAsp34Glu) in the *PNPLA1* gene. Upper panel (a) represents nucleotide sequence in a homozygous affected member, middle panel (b) a heterozygous carrier and lower panel (c) a homozygous unaffected member. (d) Schematic representations showing the coding regions of patatin-like phospholipase domain-containing protein 1 isoform 2 (NM_173676.2). The * represents a variant site in exon 2. (e) Predicted structure of PNPLA1 proteins including the patatin domain beginning at Ile16, and hydrophobic domain lies between Leu336 and Ser418, and end of the protein at Gln532. Arrows indicate three previously reported (p.Ala59Val, p.Glu131*, p.Ala34Thr) and a novel mutation p.Asp34Glu (this report) in the patatin domain. (f) Three-dimensional structure of the patatin domain indicating beta sheets in yellow, alpha helices in red, and wild type polar charged Asp34 in green colour. The panel (g) indicates mutant Glu34 in light green colour. (h) Computed surface of the patatin-like domain, coloured by hydrophobicity. (i) Comparison of partial amino acid sequence of human PNPLA1 across different species. Aspartate (D) within the box indicates the conserved residue across different species. The missense mutation (p.Asp34Glu) affecting conserved aspartate residue in human PNPLA1 is indicated by an arrow.

Table 1

List of mutations reported in the *PNPLA1* gene so far

Nucleotide variant	Amino acid change	Mutation type	Reported previously by	PolyPhen-2.1 (HumVar)†				SIFT‡	
				Score	Sensitivity	Specificity	Prediction	Score	Prediction
c.102C>A	p.Asp34Glu	Missense	This report	0.983	0.56	0.94	Probably damaging	0.012	Damaging
c.100G>A	p.Ala34Thr	Missense	Fachal *et al.*[7]	0.979	0.57	0.94	Probably damaging	0.01	Damaging
c.176C>T	p.Ala59Val	Missense	Grall *et al.*[3]	1	0	1	Probably damaging	0.02	Damaging
c.391G>T	p.Glu131*	Nonsense	Grall *et al.*[3]	–	–	–	–	–	–

†PolyPhen-2.1 (http://genetics.bwh.harvard.edu/pph2). Used in default mode with native alignment.

‡SIFT (http://sift.jcvi.org). Used in default mode with native alignment.

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