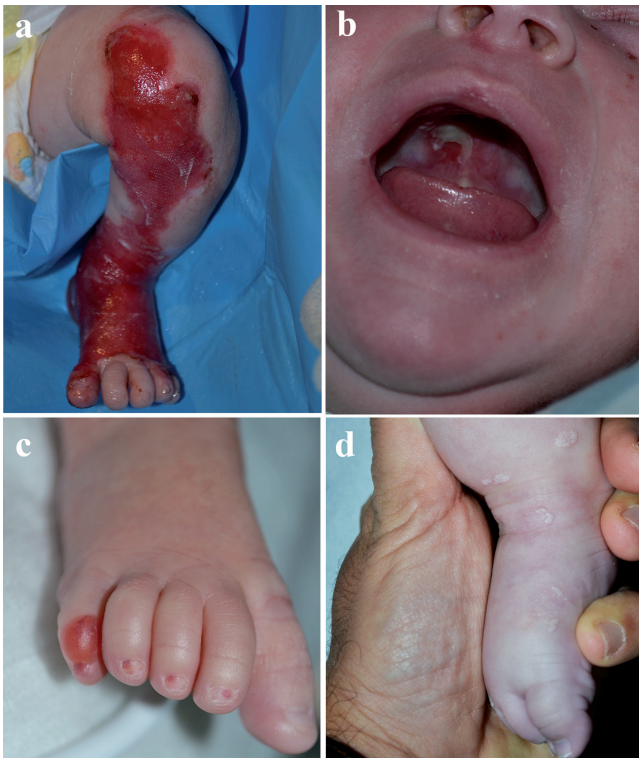

Fig. S1. Case 2: (a) aplasia cutis congenita of the left leg and (b) palate erosions at 7 days of age. Case 4: (c) haemorrhagic blister on the fifth toe and subungual haemorrhages in the second day of life, while only numerous milia (d) at sites of healed lesions are present at the 3-month follow-up visit.

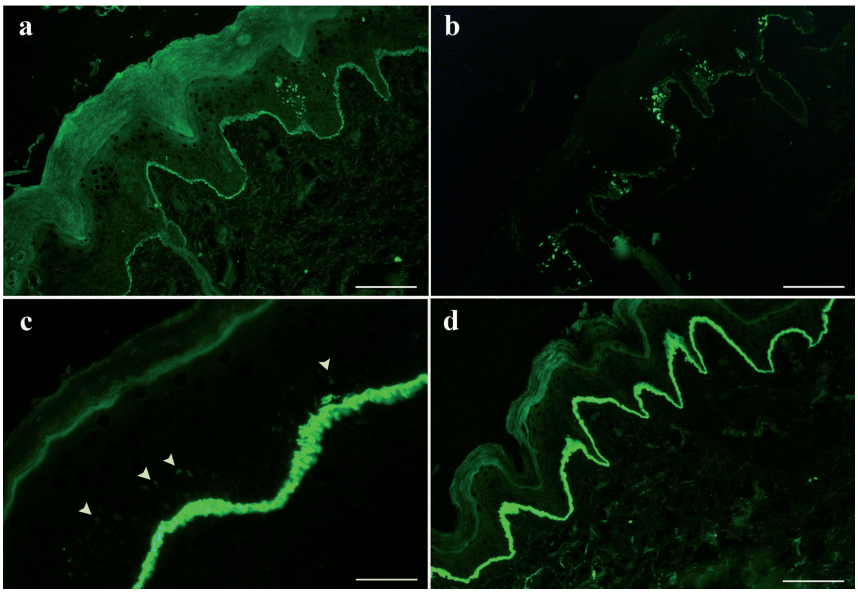

Fig. S2. Immunofluorescence antigen mapping with anti-type VII monoclonal antibody shows granular deposits irregularly distributed within the epidermis in (a) patient 3 and (b) patient 4, while the linear labelling along the cutaneous basement membrane zone (BMZ) appears reduced compared with (d) normal control skin. In skin sections from the father of patient 4 a faint cytoplasmic staining (*arrowheads*) is visible in some keratinocytes, while the labelling intensity along (c) the BMZ appears comparable to normal skin. *Bars*: (a, b, d) 100 µm and (c) 50 µm.

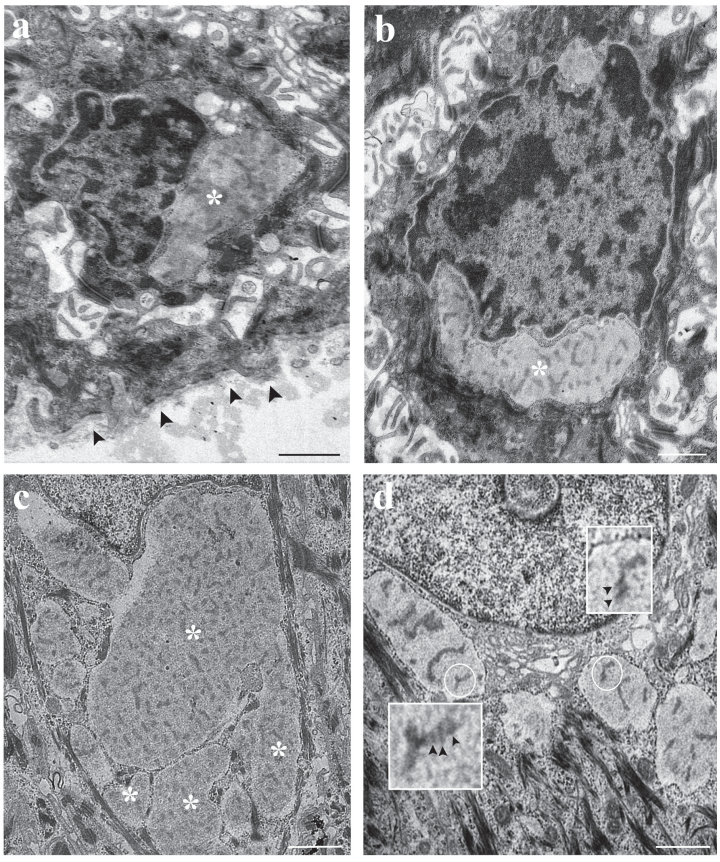

Fig. S3. Ultrastructural examination shows perinuclear cytoplasmic inclusions of variable size (*asterisks*) bounded by rough endoplasmic reticulum within basal keratinocytes of patients (a) 1, (b) 3, and (c, d) 4. Numerous roundish, elongated or branched dense bodies, corresponding to stellate bodies, are visible within the inclusions. Some dense bodies present a cross-banded pattern (d, *arrowheads* in the inset). In patient 1 a cleavage below the lamina densa (a, *arrowheads*) of the cutaneous basement membrane zone is also visible. Bars: (a–c) 2.5 µm and (d) 1.5 µm.

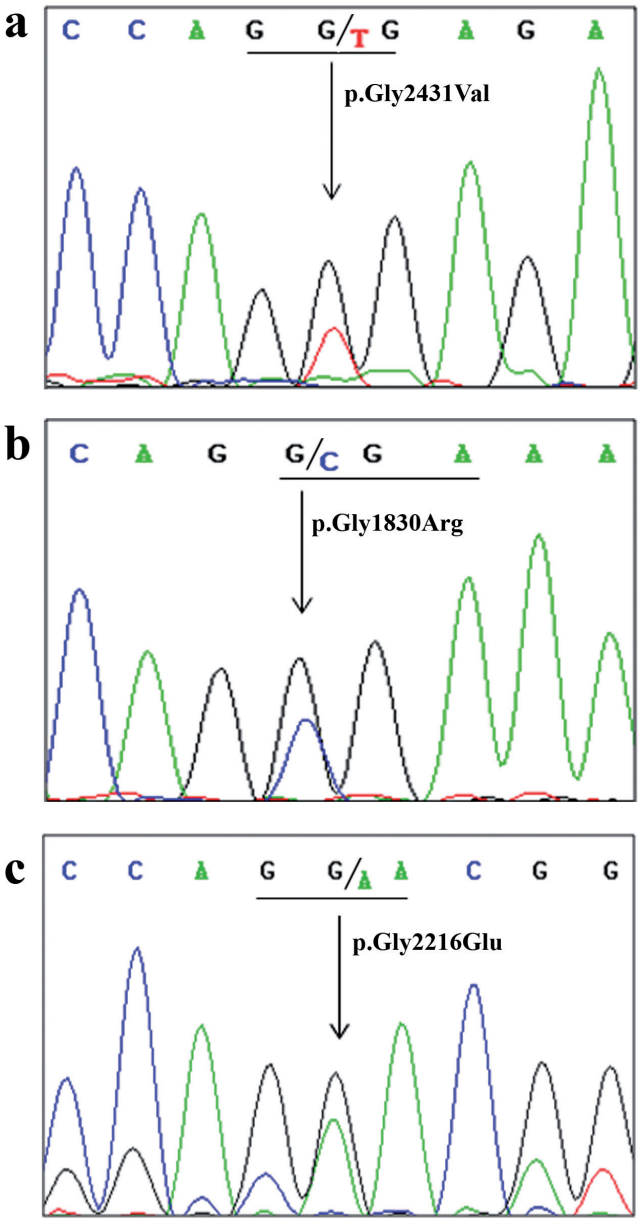

Fig. S4. Sequence chromatograms of *COL7A1* amplicons showing the novel glycine substitution mutations identified in patients with bullous dermolysis of the newborn (BDN): (a) p.Gly2431Val (dominant) in case 3, (b) p.Gly1830Arg (dominant) in case 4 and (c) p.Gly2216Glu (recessive) in case 1. The affected codon is underlined. Genotype-phenotype correlations in BDN are elusive. In our recessive cases, mutations c.4783-1G>A and c.497dupA combine with p.Pro1699Leu and p.Gly2216Glu, respectively. Both the c.4783-1G>A and c.497dupA are null mutations. The p.Pro1699Leu was previously reported in compound heterozygosity with a different splice site null mutation in an adult patient with the pretibial subtype of recessive dystrophic epidermolysis bullosa (DEB) and presenting keratinocyte cytoplasmic deposits of collagen VII in the skin (37). A similar mutation, p.Pro2259Leu, was recently described in another recessive BDN patient (see reference 28 in Table SI[1]). It remains to be determined, however, why the p.Pro1699Leu mutation may result in different DEB subtypes. Interestingly, Murase et al. reported a family with dominant DEB due to the p.Gly2242Glu mutation (see reference 26 in Table SI[1]). In this family the disease manifested as BDN in an infant and DEB pruriginosa in his mother. These examples underscore the impact of modifier genes or environmental factors in inter- and intra-familial clinical DEB variability. Glycine substitution mutations can have different degrees of severity and result in either dominant or recessive inheritance (38). In addition, the same glycine substitution may behave as a dominant mutation or be silent within the same nuclear family (see reference 24 in Table SI[1]).

Appendix S1.

MATERIALS AND METHODS

Patient samples

Following written informed consent, skin biopsies were collected from the patients and the father of patient 4. Blood samples were also obtained from the patients, their parents and the brother of patient 4. The study was conducted in compliance with the principles of the Declaration of Helsinki.

Immunofluorescence and electron microscopy studies

Frozen 5 μm thick sections were obtained from skin biopsies of the patients, patient's 4 father and healthy controls and processed for indirect immunofluorescence using the following primary antibodies: monoclonal anti-human type VII collagen (LH7.2, Sigma Immunochemical, St Louis, MO, USA); monoclonal anti-human laminin-332 (clone GB3, a gift of Dr Meneguzzi, Inserm, Nice, France); monoclonal anti-human BP180 (clone 1A8C, a gift of Dr Owaribe, Nagoya University, Japan); monoclonal anti-human integrin β4, (clone 3E1, Chemicon, Merck Millipore, Billerica, MA, USA).

For electron microscopy examination, skin biopsy specimens from patients 1–4 were fixed in 2% glutaraldehyde, post-fixed in 1% osmium tetroxide, dehydrated in graded alcohols, and embedded in Epon resin. Ultrathin sections were stained with uranyl acetate and lead citrate and observed in an Omega Zeiss EM 912 transmission electron microscope.

Molecular analysis

Mutational screening of *COL7A1* gene was performed by denaturing high-performance liquid chromatography (DHPLC) scanning of PCR products corresponding to the entire coding region, as described (7). Sanger sequencing of positive DHPLC amplicons was used to precisely identify the mutation. Each mutation was confirmed by a second cycle of PCR and sequencing.

Literature review

A literature (English, French, Italian, German and Spanish) search was performed on PubMed (http://www.ncbi.nlm.nih.gov/pubmed) from 1985 to July 2015. The following key words were used: transient bullous dermolysis of the newborn, bullous dermolysis of the newborn, dermolysis and newborn, dystrophic epidermolysis bullosa and mutation, dystrophic epidermolysis bullosa and newborn. Studies describing clinical and laboratory findings of individual BDN cases were assessed. In addition, articles reporting mutational analysis of DEB case series were examined for the inclusion of BDN cases. Finally, the International Dystrophic Epidermolysis Bullosa Patient Registry was searched for BDN cases (8). Relevant information was extracted from each article and reviewed to eliminate potential duplication of case reports.

Statistical analysis

The association between specific clinical features (congenital skin defects and mucosal involvement) and type of inheritance (dominant vs. recessive) was studied through the Fisher exact test. Statistical significance was set at the 0.05 level.

Table SI. *Clinical and molecular findings in patients with bullous dermolysis of the newborn*

				Disease extent				
Reference	Sex	Inheri-tance	Congenital skin defects	Skin	Oral mucosa	Nails	Age at skin fragility resolution	*COL7A1* mutations[a]
Hashimoto et al. (1)	M	NK	No	Generalized	No	No	12 months	NK
Hashimoto et al. (9)	M	NK	No	Generalized	No	No	17 months	NK
	F	NK	No	Generalized	No	No	6 weeks	NK
Fine et al. (5)	M	AD[b]	No	Generalized	NR	No	3 months	NK
	M	NK	No	Generalized	NR	Yes	NK (mild disease activity at 1.5 years)	NK
Fine et al. (5); Fine et al. (10)	M	NK	No	Generalized	NR	Yes	6 months	NK
	M	AD[b]*	No	Generalized	NR	No	3 months	NK
Fine et al. (10)	NR	AD[b]*	NR	NR	NR	NR	3 months	NK
	F	AD[b]**	No	Generalized	Yes	Yes	NK (mild disease activity at 6 months)	NK
	F	AD[b]**	No	Generalized	NR	Yes	6 months	NK
	M	AD***	No	Generalized	Yes	Yes	NK (residual disease activity at 13 months)	c.4120-1G>C (heterozygous)[c]
	M	AD***	No	NK	NK	Yes	NK (no blisters at 23 years)	c.4120-1G>C (heterozygous)[c]
	F	AD***	No	NK	NK	NR	NK (mild disease activity at 11 years)	c.4120-1G>C (heterozygous)[c]
	F	AD***	No	NK	NK	Yes	NK (no blisters at 37 years)	c.4120-1G>C (heterozygous)[c]
Smith & Sybert (6)	M	AR***	No	Generalized	Yes	NR	NA (died at 10 days, cardio-pulmonary arrest)	NK
McCollough et al. (11)	F	AD[b]	No	Generalized	Yes	Yes	NK (mild disease activity at 6 months)	NK
Eng et al. (12)	F	NK	No	Generalized	No	No	4 weeks	NK
Patrizi et al. (13)	F	AR[b]	Yes (lower arms and feet)	Generalized	No	No	18 months	NK
Phillips et al. (14)	F	NK	No	Generalized	NR	Yes	NK (mild disease activity at 6 months)	NK
Okuda et al. (15)	F	NK	No	Generalized (a few lesions)	No	No	1 month	NK
Hatta et al. (16)	F	NK	Yes (right leg and knee and feet)	Generalized	Yes	Yes	NK (residual disease activity at 4 years)	NK
D'incan et al. (17)	F	NK	No	Generalized	No	No	3 weeks	NK
Hammami-Hauasli et al. (4)	F	AR	Yes (lower legs)	Generalized	NR	Yes	NK (mild disease activity at 14 months)	p.Gly1519Asp/p.Gly2251Glu
Hanson et al. (18)	M	NK	No	Localized (acral)	NR	NR	1 month	NK
	F	NK	Yes (from knees to toes)	Generalized	Yes	NR	NK (residual disease activity at 7 months)	NK
	M	NK	No	Localized (acral)	Y	NR	2 months	NK
Fassihi et al. (19)	M	AD	No	Localized (acral)	No	No	4 months	p.Gly1522Glu (heterozygous)
Nakano et al. (20)	M	AR	Yes (first toes)	Generalized	Yes	Yes	29 months	p.Arg990Gln in linkage with c.5504delA/p.Arg2008His
Oh et al. (21)	F	AR	Yes (right lower leg)	Localized (acral)	NR	NR	2 months	p.Gly798Arg/c.6246del27
Hashikawa et al. (22)	M	AR	No	Generalized	NR	Yes	12 months	c.682+1G>A/p.Gly1910Ser
Oppenheimer & Hallas (23)	F	AR[b]	No	Generalized	No	NR	NK (residual disease activity at 3 months)	NK
Almaani et al. (24)	F	AD	No	Generalized	NR	NR	4 months	p.Gly1483Asp (heterozygous)
Frew et al. (25)	M	AD	No	Generalized	No	NR	18 months	p.Gly1673Arg (heterozygous)
Murase et al. (26)	M	AD	No	Localized (acral)	No	No	6 months	p.Gly2242Glu (heterozygous)
Radkevich-Brown &	M	AR[b]	Yes (right foot)	Generalized	Yes	Yes	7 months	NK
Schwayder (27)	M	AR[b]	Yes (left foot)	Localized (acral)	Yes	Yes	4 months	NK
	M	NK	No	Generalized	NR	NR	3 months	NK
	M	NK	No	Generalized	Yes	Yes	5 months	NK
Boccaletti et al. (28)	F	AR	No	Generalized	Yes	Yes	NK (mild disease activity at 3 years)	p.Pro2259Leu (homozygous)
	M	AR	No	Generalized	Yes	Yes	NK (minimal disease activity at 10 months)	p.Pro2259Leu (homozygous)
Shi et al. (29)	M	AD	Yes (pretibial areas)	Localized (legs, feet)	No	No	2 months	p.Gly2046Ser
Current study								
1	F	AR	Yes (from knees to feet)	Generalized	Yes	Yes	3 months	c.497dupA/p.Gly2216Glu
2	M	AR	Yes (left leg, from knee to foot)	Localized (lower extremities and face)	Yes	No	3 months	c.4783-1G>A/ p.Pro1699Leu
3	M	AD	No	Localized (extremities and face)	No	Yes	Residual disease activity at 3 years	p.Gly2431Val
4	M	AD	No	Generalized	No	Yes	3 months	p.Gly1830Arg

At least 4 additional molecularly characterized cases were reported, but were not included in the analysis because of insufficient clinical description (24, 30).
[a]Mutation designation is based on the cDNA sequence deposited under the GenBank entry NM_000094.3. [b]Inheritance based on suggestive family history. [c]The mutation in this family has been identified and described by Christiano et al. (3). *Members of family 2 (10); **members of family 3 (10); ***members of family 4 (10).
NR: not reported; NK: not known; NA: not applicable.