Hernández-Martín, A., Cuadrado-Corrales, N., Ciria-Abad, S., Arias-Palomo, D., Mascaró-Galy, J., Escámez, M., García, M., del Río, M., Torrelo, A. & González-Sarmiento, R. (2010). X-Linked Ichthyosis along with Recessive Dystrophic Epidermolysis Bullosa in the Same Patient. *Dermatology, 221*(2), pp. 113–116.

DOI: 10.1159/000313507

X-Linked Ichthyosis along with Recessive Dystrophic Epidermolysis Bullosa in the Same Patient

A. Hernández-Martín[a] N. Cuadrado-Corrales[b] S. Ciria-Abad[e] D. Arias-Palomo[c]
J.M. Mascaró-Galy[d] M.J. Escámez[b] M. García[b] M. Del Río[b] A. Torrelo[a]
R. González-Sarmiento[e]

[a]Department of Dermatology, Hospital Niño Jesús, [b]CIEMAT-CIBERER U714, Basic Research Department, Epithelial Biomedicine Division, Regenerative Medicine Unit, and [c]Department of Dermatology, Hospital Fuelanbrada, Madrid; [d]Department of Dermatology, Hospital Clínic i Provincial, Barcelona; [e]Molecular Medicine Unit, Department of Medicine and IBMCC, University of Salamanca and CSIC, Salamanca, Spain

Key Words
X-linked ichthyosis · Epidermolysis bullosa · Genodermatoses

Abstract
X-linked ichthyosis (XLI) is a relatively common keratinization disorder which is caused, in the vast majority of cases, by a total deletion of the sulfatase steroid (STS) gene. Dystrophic epidermolysis bullosa (DEB) is a scarring form of epidermolysis bullosa of either autosomal recessive or dominant inheritance secondary to collagen VII gene mutations. We report the first case of a patient with both XLI and DEB in whom a partial deletion of the STS gene and a recessive point mutation in *COL7A1* were demonstrated.

Introduction

X-linked ichthyosis (XLI) is a fairly common keratinization disorder caused by a sulfatase steroid (STS) deficiency. In most XLI cases, the STS gene is totally deleted, as can easily be confirmed by PCR techniques. Dystrophic epidermolysis bullosa (DEB) is due to *COL7A1* gene mutations, and has been mapped to chromosome 3. It is inherited as an autosomal recessive or dominant trait. We report the first case of both XLI and DEB in a single patient in which a partial deletion of the STS gene and a recessive mutation in *COL7A1* were demonstrated.

Case Report

An 8-year-old boy was referred to our clinic because of extreme skin dryness present since the first year of life. The child's parents were non-consanguineous, and he was born by C-section after a normal pregnancy. His younger and sole brother, 2 maternal uncles, and his maternal grandfather also suffered from 'dry skin'. The patient had been diagnosed with DEB since birth and, according to his medical record, he was born with extensive erosive areas on the right leg and feet (fig. 1). A skin biopsy performed at birth showed subepidermal blistering, but no further ultrastructural or genetic tests were carried out at that time. There was no family history of cutaneous blistering.

On physical examination, the patient had large polygonal, dark-brown scales on the extensor aspects of the limbs, as well as blisters and scars over the bony prominences of the knees, ankles, dorsal areas of the hands and feet, elbows and spine (fig. 2). There were absent nails on several fingers and toes (fig. 3). The younger brother also showed dark-brown polygonal scales on the abdomen and the extensor areas of the arms and legs. We could not examine the rest of the family. Genomic DNA from both the children and their parents was extracted from peripheral blood lymphocytes by standard methods, and molecular analysis of the exons 1, 5 and 10 of the STS gene by PCR was performed [1–3]. The PCR test showed no DNA amplification of exons 5 and 10, thus confirming a partial deletion of the STS gene at the 3′ end in both siblings. No other relatives were available for STS molecular testing. To better characterize the blistering process in the elder brother, immunomapping of a perilesional skin biopsy with a battery of antibodies against all the dermo-epidermal proteins known to be altered in different types of epidermolysis bullosa was performed, yielding no expression abnormalities. Unfortunately, we were unable to identify blisters at the dermo-epidermal junction. It is worth noting that immunomapping is a semi-quantitative test, so in mild clinical presentations some mutated proteins may show a normal

Angela Hernández-Martín, MD
Department of Dermatology, Hospital Niño Jesús
Avda. Menendez Pelayo, 65
ES–28009 Madrid (Spain)
E-Mail ahernandez_hnj@yahoo.es

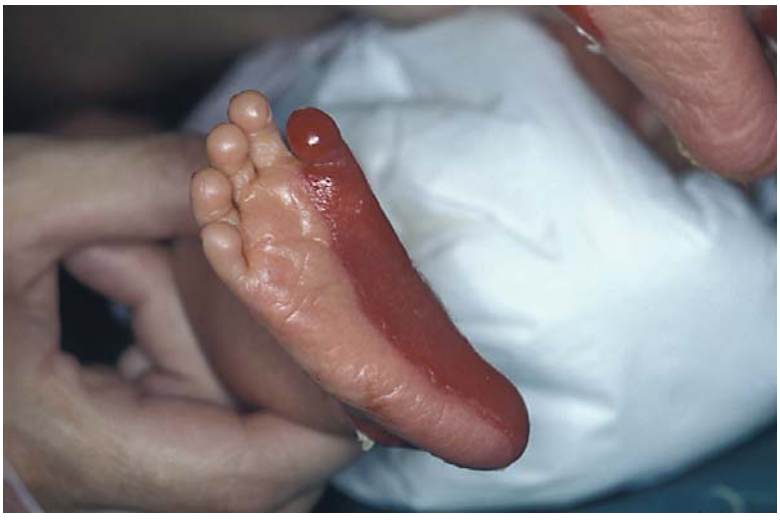

Fig. 1. Clinical aspect at birth. Large absence of the skin on the right foot.

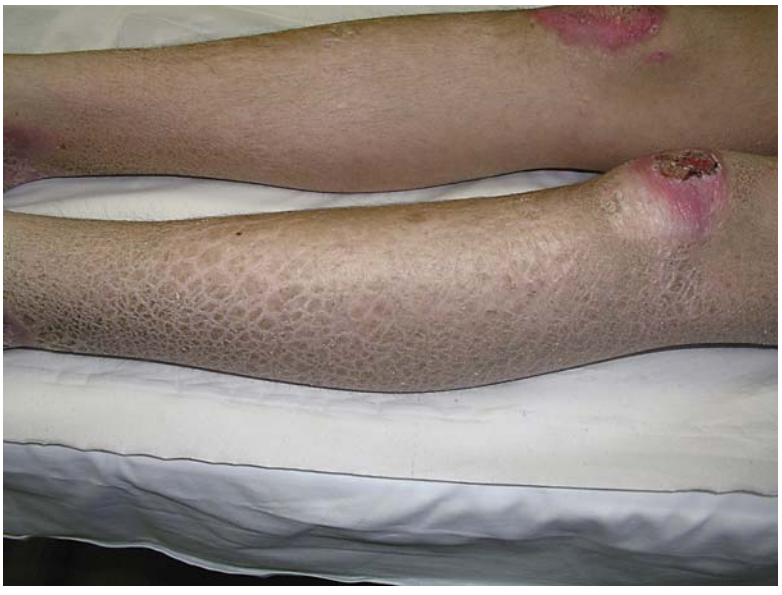

Fig. 2. Dark-brown polygonal scales on the extensor aspects of both legs typical of XLI. Erosions, crusts and dystrophic scars on the knees, a typical feature of DEB.

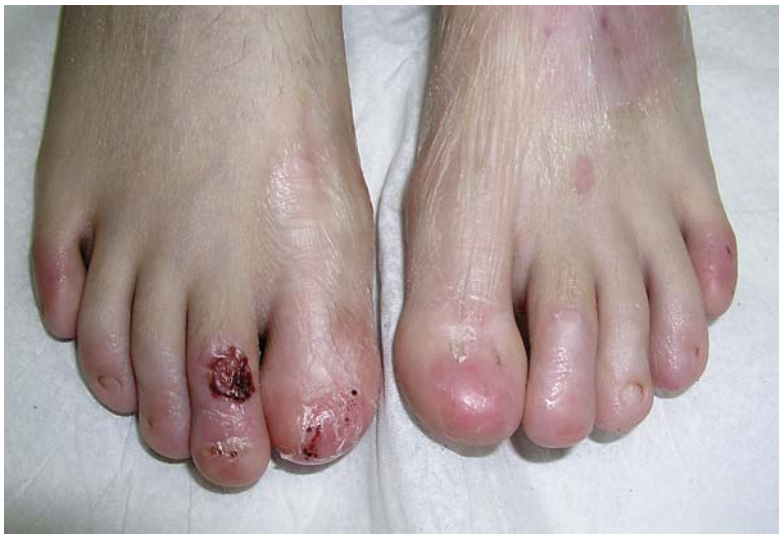

Fig. 3. Missing and dystrophic toenails as well as hemorrhagic blisters and crusts on the fingers.

level of expression. Transmission electron microscopy was not performed. Despite the fact that these preliminary studies were not conclusive, we decided, based on the clinical phenotype, that the candidate gene to be screened was the *COL7A1* gene. The entire coding sequence of the *COL7A1* gene (118 exons), flanking intron boundaries and the promoter were amplified by touchdown PCR with the primer pairs previously described [4, 5]. Amplicons were sequenced with ABI Prism 3730 (Applied Biosystems) and screened for mutation. This study disclosed a heterozygous c.6527insC mutation in the DNA from the patient. In addition, the father was a healthy carrier of the mutation. However, a most probable mutation in the other allele could not be found. We have currently lost contact with the patients. Throughout the follow-up period, the scaling condition of the younger child improved due to the regular use of topical keratolytics and emollients, but it was still quite noticeable in the elder brother, as he has been taught to avoid any kind of mechanical rubbing of the skin and he did not like applying moisturizers or washing the skin with a foam. Recurrent DEB blisters were treated with either topical antiseptic or antibiotics.

Discussion

XLI is caused by STS deficiency. Within the epidermis, this STS deficiency leads to elevated cholesterol sulphate levels in the stratum corneum, altering the physical properties of the corneocyte membrane and increasing their stability and degree of intercellular cohesion [6]. The condition appears early in life, and is characterized by large adherent scales of a brownish hue on different parts of the body surface, more evident on the extensor surfaces of the extremities. Diagnosis of patients with XLI is based on either demonstration of an STS deficiency or an STS gene abnormality [7]. The STS gene has been mapped to the distal part of the short arm of the X chromosome (Xp21.3). The Y chromosome does not have a functional STS gene, although a pseudogene of 100 kb is known to exist on the long arm of the Y chromosome [8]. In X-STS gene only exons 1, 5 and 10 had non-homologous nucleotide sequences [9]. Therefore, PCR studies to detect STS gene deletions in males can be performed exclusively with such exons. 90% of patients with XLI show a complete deletion of the STS gene, while a minority of cases display partial deletions [10–13] or point mutations in the nucleotide sequence [14–16]. In our cases, PCR tests showed a partial deletion of the 3′ end of the STS gene involving exons 5 and 10 in both children, a genetic anomaly occurring in less than 10% of patients suffering from XLI. We could not access other male members of the maternal pedigree complaining of 'dry skin', but they most probably had the same genetic abnormality.

Hereditary epidermolysis bullosa is a severe genodermatosis characterized by skin fragility after a minor trauma. Although there is an isolated report of a patient suffering from both epidermolysis bullosa simplex and XLI [17], the association of a hereditary epidermolysis bullosa and XLI seems to be exceptional. DEB is a scarring form of epidermolysis bullosa in which there is a dermoepidermal cleavage under the lamina densa of the basement membrane. The pattern of inheritance may be either autosomal recessive or autosomal dominant, both forms of DEB being caused by mutations of the *COL7A1* gene [18]. This gene spans over approximately 32 kb of the 3p21.1 region and contains 118 exons [19, 20]; it encodes collagen type VII, the main component of the anchoring fibrils at the dermo-epidermal junction. Clinical findings in DEB range from a generalized severe mucocutaneous blistering and scarring, to scarce bulla and scars localized solely over the bony prominences. Because there is a significant clinical overlap between the localized mild autosomal recessive type and the dominant form of DEB – and because ultrastructural studies are not always conclusive – precise diagnosis may require molecular analysis in the affected individual and his/her parents [21]. Several studies have shown these clinical manifestations to depend on the type of *COL7A1* gene mutation [22, 23]; while mild autosomal recessive forms derive from a variety of mutations leading to the expression of altered but partially functioning collagen VII molecules [24], the dominant mutations are typically caused by glycine substitutions in the *COL7A1* gene promoting a dominant-negative interference with the wild-type protein [25]. It is known that most cases of mild-to-moderate severity correspond to recessive mutations [23, 26], but genetic testing is mandatory for an accurate genetic counseling. In our patient, this study disclosed the presence of the c.6527insC mutation in a single allele, most probably in combination with a still unidentified second mutation. The pathogenic effect of c.6527insC was well established in the present study, as well as in 2 previous reports [27, 28]. The insertion of C6527 in exon 80 creates a shift in the reading frame resulting in a TAA stop codon 337 bp downstream of codon 2176. The absence of collagen VII immunolabeling and of anchoring fibrils in homozygous patients supports the hypothesis of an in vivo down-regulation of the mutant transcript by the nonsense-mediated mRNA decay system shortly after their transcription [29, 30]. This c.6527insC mutation has been previously reported as a recessive null recurrent mutation in the Spanish RDEB patients accounting for 47.5% of RDEB alleles of a cohort of 41 patients [31]. The presence of this mutation is compatible with a DEB inherited in a recessive manner together with a second mutation not yet identified. Screening by direct sequencing in our group has a detection sensitivity of 93.3% of all *COL7A1* mutations in DEB [31]. However, some larger internal deletion or insertions, branch-point, or other non-coding gene or RNA abnormalities may be missed.

Our patient had a mild recessive form of DEB, but he developed a most noticeable XLI phenotype because he had been taught not to rub the skin under any circumstances. As the scaling was cosmetically well tolerated by the patient, at that time we did not consider treatment with oral retinoids, but recommended that he should gently apply hydrating agents and moisturizers to facilitate the elimination of the scales without inducing skin detachment.

This is the first report of a patient with both XLI and DEB, two genetically unrelated disorders that are most probably randomly associated here. We demonstrated a partial deletion of the *STS* gene together with the c.6537insC recessive mutation in the *COL7A1* gene.

Acknowledgment

We gratefully acknowledge Prof. John McGrath and his team for their help in genetic testing.

References

1 Nomura K, Nakano H, Umeki K, Harada K, Kon A, Tamai K, et al: A study of the steroid sulfatase gene in families with X-linked ichthyosis using polymerase chain reaction. Acta Derm Venereol 1995;75:340–342.

2 Sugawara T, Iwaki M, Fujimoto S: Diagnosis of a deletion of steroid sulfatase by polymerase chain reaction and high-performance liquid chromatography. Clin Chim Acta 1997;263:25–32.

3 Aviram-Goldring A, Goldman B, Netanelov-Shapira I, Chen-Shtoyerman R, Zvulunov A, Tal O, et al: Deletion patterns of the STS gene and flanking sequences in Israeli X-linked ichthyosis patients and carriers: analysis by polymerase chain reaction and fluorescence in situ hybridization techniques. Int J Dermatol 2000;39:182–187.

4 Christiano AM, Hoffman GG, Zhang X, Xu Y, Tamai Y, Greenspan DS, et al: Strategy for identification of sequence variants in *COL7A1* and a novel 2-bp deletion mutation in recessive dystrophic epidermolysis bullosa. Hum Mutat 1997;10:408–414.

5 Gardella R, Barlati S, Zoppi N, Tadini G, Colombi M: A –96C–>T mutation in the promoter of the collagen type VII gene *(COL7A1)* abolishing transcription in a patient affected by recessive dystrophic epidermolysis bullosa. Hum Mutat 2000;16:275.

6 Williams ML: Epidermal lipids and scaling diseases of the skin. Semin Dermatol 1992; 11:169–175.

7 Hernandez-Martin A, Gonzalez-Sarmiento R, De Unamuno P: X-linked ichthyosis: an update. Br J Dermatol 1999;141:617–627.

8 Yen PH, Marsh B, Allen E, Tsai SP, Ellison J, Connolly L, et al: The human X-linked steroid sulfatase gene and a Y-encoded pseudogene: evidence for an inversion of the Y chromosome during primate evolution. Cell 1988;55:1123–1135.

9 Ballabio A, Ranier JE, Chamberlain JS, Zollo M, Caskey CT: Screening for steroid sulfatase (STS) gene deletions by multiplex DNA amplification. Hum Genet 1990;84:571–573.

10 Ballabio A, Carrozzo R, Parenti G, Gil A, Zollo M, Persico MG, et al: Molecular heterogeneity of steroid sulfatase deficiency: a multicenter study on 57 unrelated patients, at DNA and protein levels. Genomics 1989;4: 36–40.

11 Bonifas JM, Morley BJ, Oakey RE, Kan YW, Epstein EH Jr: Cloning of a cDNA for steroid sulfatase: frequent occurrence of gene deletions in patients with recessive X chromosome-linked ichthyosis. Proc Natl Acad Sci USA 1987;84:9248–9251.

12 Shapiro LJ, Yen P, Pomerantz D, Martin E, Rolewic L, Mohandas T: Molecular studies of deletions at the human steroid sulfatase locus. Proc Natl Acad Sci USA 1989;86:8477–8481.

13 Yen PH, Marsh B, Allen E, Tsai SP, Ellison J, Connolly L, et al: The human X-linked steroid sulfatase gene and a Y-encoded pseudogene: evidence for an inversion of the Y chromosome during primate evolution. Cell 1988;55:1123–1135.

14 Alperin ES, Shapiro LJ: Characterization of point mutations in patients with X-linked ichthyosis: effects on the structure and function of the steroid sulfatase protein. J Biol Chem 1997;272:20756–20763.

15 Basler E, Grompe M, Parenti G, Yates J, Ballabio A: Identification of point mutations in the steroid sulfatase gene of three patients with X-linked ichthyosis. Am J Hum Genet 1992;50:483–491.

16 Gonzalez-Huerta LM, Messina-Baas OM, Toral-Lopez J, Rivera-Vega MR, Kofman-Alfaro S, Cuevas-Covarrubias SA: Point mutation in the STS gene in a severely affected patient with X-linked recessive ichthyosis. Acta Derm Venereol 2006;86:78–79.

17 Steuhl KP, Anton-Lamprecht I, Arnold ML, Thiel HJ: Recurrent bilateral corneal erosions due to an association of epidermolysis bullosa simplex Köbner and X-linked ichthyosis with steroid sulfatase deficiency. Graefes Arch Clin Exp Ophthalmol 1988;226:216–223.

18 Burgeson RE: Type VII collagen, anchoring fibrils, and epidermolysis bullosa. J Invest Dermatol 1993;101:252–255.

19 Christiano AM, Greenspan DS, Lee S, Uitto J: Cloning of human type VII collagen: complete primary sequence of the alpha 1(VII) chain and identification of intragenic polymorphisms. J Biol Chem 1994;269:20256–20262.

20 Christiano AM, Hoffman GG, Chung-Honet LC, Lee S, Cheng W, Uitto J, et al: Structural organization of the human type VII collagen gene *(COL7A1)*, composed of more exons than any previously characterized gene. Genomics 1994;21:169–179.

21 Hashimoto I, Kon A, Tamai K, Uitto J: Diagnostic dilemma of 'sporadic' cases of dystrophic epidermolysis bullosa: a new dominant or mitis recessive mutation? Exp Dermatol 1999;8:140–142.

22 Jarvikallio A, Pulkkinen L, Uitto J: Molecular basis of dystrophic epidermolysis bullosa: mutations in the type VII collagen gene *(COL7A1)*. Hum Mutat 1997;10:338–347.

23 Uitto J, Christiano AM: Molecular basis for the dystrophic forms of epidermolysis bullosa: mutations in the type VII collagen gene. Arch Dermatol Res 1994;287:16–22.

24 Gardella R, Zoppi N, Zambruno G, Barlati S, Colombi M: Different phenotypes in recessive dystrophic epidermolysis bullosa patients sharing the same mutation in compound heterozygosity with two novel mutations in the type VII collagen gene. Br J Dermatol 2002;147:450–457.

25 Mellerio JE, Salas-Alanis JC, Talamantes ML, Horn H, Tidman MJ, Ashton GH, et al: A recurrent glycine substitution mutation, G2043R, in the type VII collagen gene *(COL7A1)* in dominant dystrophic epidermolysis bullosa. Br J Dermatol 1998;139: 730–737.

26 Hashimoto I, Kon A, Tamai K, Uitto J: Diagnostic dilemma of 'sporadic' cases of dystrophic epidermolysis bullosa: a new dominant or mitis recessive mutation? Exp Dermatol 1999;8:140–142.

27 Hovnanian A, Rochat A, Bodemer C, Petit E, Rivers CA, Prost C, et al: Characterization of 18 new mutations in *COL7A1* in recessive dystrophic epidermolysis bullosa provides evidence for distinct molecular mechanisms underlying defective anchoring fibril formation. Am J Hum Genet 1997;61:599–610.

28 Kern JS, Kohlhase J, Bruckner-Tuderman L, Has C: Expanding the *COL7A1* mutation database: novel and recurrent mutations and unusual genotype-phenotype constellations in 41 patients with dystrophic epidermolysis bullosa. J Invest Dermatol 2006;126:1006–1012.

29 Couttet P, Grange T: Premature termination codons enhance mRNA decapping in human cells. Nucleic Acids Res 2004;32:488–494.

30 Dang N, Murrell DF: Mutation analysis and characterization of *COL7A1* mutations in dystrophic epidermolysis bullosa. Exp Dermatol 2008;17:553–568.

31 Escámez MJ, Garcia M, Cuadrado-Corrales N, Llames S, Charlesworth, de Luca N, et al: The first *COL7A1* mutation survey in a large Spanish Dystrophic Epidermolysis Bullosa cohort: c.6527insC disclosed as an unusually recurrent mutation. Br J Dermatol 2010, E-pub ahead of print.