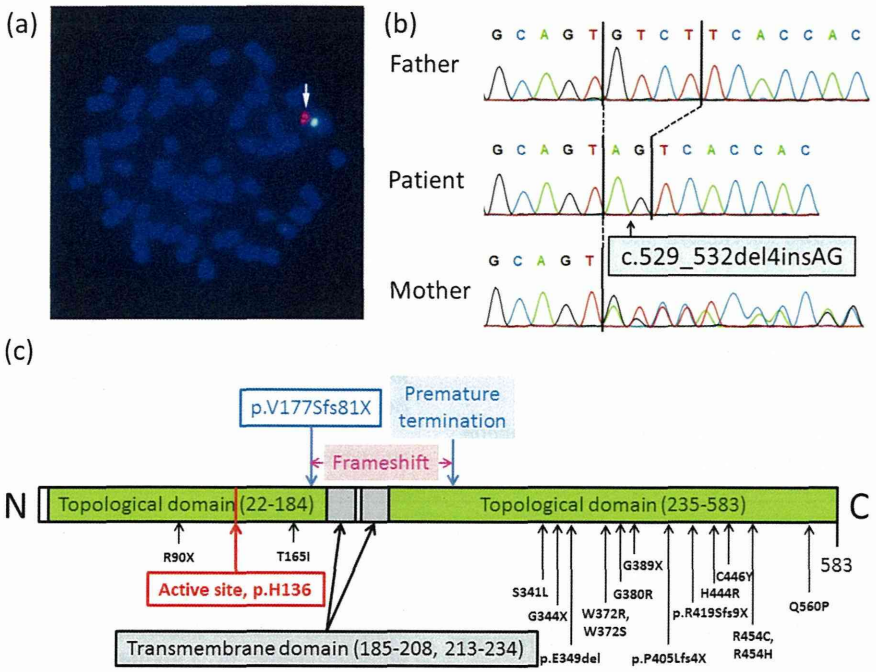


Fig. 2. FISH of Xp22.3, hemizygous mutations of STS of the patient, and the scheme of STS domain structure.
(a) Fluorescence *in situ* hybridization test for Xp22.3 indicates the existence of Xp22.3 (white arrow) in the patient. Red probe: region of *STS*, Xp22.3; green probe: a control for chromosome X. (b) Sanger sequencing reveals a hemizygous c.529_532del4insAG mutation of STS in the patient. His mother is a heterozygous carrier whereas his father shows wild-type sequence only. (c) The scheme of domain structure of STS. STS has two topological domains (green areas, amino acids 22–184, 235–583) and two transmembrane domains (gray areas, amino acids 185–208, 213–234). The N-terminal topological domain contains the reported active site, p.136 His (red arrow). Sites of the present novel indel mutation p.Val 177 Serfs × 81 in exon 5 and the premature termination are shown with blue arrows (top). Sites of the previously described mutations are indicated with black arrows (bottom).

Therefore, we hypothesize that there may be some residual STS enzyme activity in this case that contributed to the skin healing.

In summary, our case expands the phenotypic diversity and outcomes in RXLI, and highlights the value of WES in accurately identifying a pathogenic mutation in this subtype of ichthyosis, as well as providing a database for the future elucidation of other genetic modifiers contributing to the phenotypic variability.

Funding statement

None.

Conflict of interest

The authors have no conflict of interest to declare.

Acknowledgments

The authors thank Ms. Rashida Pramanik, Ms. Haruka Ozeki and Ms. Yuka Terashita for their technical help in analysing mutations of *STS*. This study was supported in part by a Grant-in-Aid for Scientific Research (B) to M.A. (15H04887), a Grant-in-Aid for Challenging Exploratory Research to M.A. (15K15415), a Grant-in-Aid for Scientific Research (B) to K.S. (15H04886) and a Grant-in-Aid for Challenging Exploratory Research to K.S. (15K15414) from the Ministry of Education, Culture, Sports, Science and Technology of Japan. The study is also supported by the UK National Institute for Health Research (NIHR) Biomedical Research Centre based at Guy's and St. Thomas' NHS Foundation Trust and King's College London.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.jdermsci.2015.07.001.

References

[1] D. Webster, J.T. France, L.J. Shapiro, R. Weiss, X-linked ichthyosis due to steroid-sulphatase deficiency, Lancet 1 (1978) 70–72.
[2] N. Hosomi, K. Fukai, A. Tanaka, H. Fujita, M. Ishii, Fluorescence *in situ* hybridization analysis is useful for the diagnosis of the carrier state of X-linked ichthyosis, Int. J. Dermatol. 47 (2008) 529–530.
[3] J.L. Hand, C.K. Runke, J.C. Hodge, The phenotype spectrum of X-linked ichthyosis identified by chromosomal microarray, J. Am. Acad. Dermatol. 72 (2015) 617–627.
[4] M. Valdes-Flores, S.H. Kofman-Alfaro, A.L. Jimenez-Vaca, S.A. Cuevas-Covarrubias, Carrier identification by FISH analysis in isolated cases of X-linked ichthyosis, Am. J. Med. Genet. A 102 (2001) 146–148.
[5] E. Basler, M. Grompe, G. Parenti, J. Yates, A. Ballabio, Identification of point mutations in the steroid sulfatase gene of three patients with X-linked ichthyosis, Am. J. Hum. Genet. 50 (1992) 483–491.
[6] A. Hernandez-Martin, R. Gonzalez-Sarmiento, P. De Unamuno, X-linked ichthyosis: an update, Br. J. Dermatol. 141 (1999) 617–627.
[7] G. Murtaza, S. Siddiq, S. Khan, S. Hussain, M. Naeem, Molecular study of X-linked ichthyosis: report of a novel 2-bp insertion mutation in the STS and a very rare case of homozygous female patient, J. Dermatol. Sci. 74 (2014) 165–167.
[8] T. Takeichi, L. Liu, K. Fong, L. Ozoemena, J.R. McMillan, A. Salam, et al., Whole-exome sequencing improves mutation detection in a diagnostic epidermolysis bullosa laboratory, Br. J. Dermatol. 172 (2015) 94–100.
[9] F.G. Hernandez-Guzman, T. Higashiyama, W. Pangborn, Y. Osawa, D. Ghosh, Structure of human estrone sulfatase suggests functional roles of membrane association, J. Biol. Chem. 278 (2003) 22989–22997.

Takuya Takeichi[a,b]
[a]*Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan*; [b]*St. John's Institute of Dermatology, King's College London, Guy's Hospital, Great Maze Pond, London SE1 9RT, UK*

Kazumitsu Sugiura[a]
[a]*Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan*

Chao-Kai Hsu[b,c]
[b]St. John's Institute of Dermatology, King's College London, Guy's Hospital, Great Maze Pond, London SE1 9RT, UK; [c]Department of Dermatology, National Cheng Kung University Hospital, College of Medicine, National Cheng Kung University, Tainan, Taiwan

Kana Tanahashi[a]
[a]Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan

Hiroyuki Takama[a]
[a]Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan

Michael A. Simpson[d]
[d]Division of Genetics and Molecular Medicine, King's College London, Guy's Hospital, Great Maze Pond, London SE1 9RT, UK

John A. McGrath[b]
[b]St. John's Institute of Dermatology, King's College London, Guy's Hospital, Great Maze Pond, London SE1 9RT, UK

Masashi Akiyama[a,*]
[a]Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan

*Corresponding author. Fax: +81 52 744 2318
E-mail address: makiyama@med.nagoya-u.ac.jp (M. Akiyama).

Received 28 May 2015
Received in revised form 24 June 2015
Accepted 1 July 2015

http://dx.doi.org/10.1016/j.jdermsci.2015.07.001

Letter to the Editor

Impetigo herpetiformis with *IL36RN* mutations in a Chinese patient: A founder haplotype of c.115+6T>C in East Asia

Keywords:
Founder haplotype
Generalized pustular psoriasis
IL36RN
Impetigo herpetiformis
Interleukin-36 receptor antagonist

Impetigo herpetiformis (IH) is a rare pustular dermatosis of pregnancy [1]. Most patients with IH do not have personal and family histories of psoriasis. Early diagnosis is essential, as IH occasionally leads to maternal or foetal death. Despite the clinical importance of IH, its aetiology has not been clarified sufficiently. Recently, we reported two Japanese cases of IH with homozygous and heterozygous mutations of *IL36RN*, which encodes the interleukin-36 receptor antagonist (IL-36RN) [2]. However, the incidence of IH cases with *IL36RN* mutations is unknown. To our knowledge, no subsequent case of IH with or without an *IL36RN* mutation has been reported thus far.

After a long-standing controversy over whether IH is an independent disease entity from generalized pustular psoriasis (GPP), today there is a tentative consensus that IH is GPP occurring during pregnancy [3]. We reported that most GPP cases that are not accompanied by psoriasis vulgaris (PV; GPP alone) are caused by *IL36RN* mutations, although only a small number of cases with GPP preceding or accompanied by PV were found to have *IL36RN* mutations [4].

Here, we report a case of IH in a Chinese patient with a homozygous *IL36RN* mutation c.115+6T>C, the most frequent GPP-causing mutation in the Chinese population. We also found a novel haplotype of *IL36RN* c.115+6T>C, which is a probable founder haplotype, both in the present patient and in 2 Japanese families.

The patient was a 25-year-old Chinese woman who was admitted to our hospital for pustular lesions after her first normal vaginal delivery (Fig. 1a). She had neither a family history of GPP and IH nor consanguinity in her family. She had no history of GPP. Her pustular lesions began to develop at the 29th week of her first pregnancy, and she had been treated in a maternity hospital. Oral betamethasone of 3 mg/day was administrated, although the eruptions persisted. The skin biopsy of a specimen from a pustular eruption on the trunk revealed a spongiform pustule of Kogoj in the epidermis, consistent with IH (Fig. 1b). She had erythema with pustules all over her body and fever of a body temperature higher than 38 °C. Blood examinations revealed white blood cell counts of 31,590/μL and C-reactive protein concentrations of 2.45 mg/dL (reference range: <0.3 mg/dL). Bacterial culture of the pustules yielded negative results. Thus, she was diagnosed as having IH.

After ethical approval, informed consent was obtained in compliance with the guidelines of the Declaration of Helsinki. The entire coding regions of *IL36RN*, including the exon/intron boundaries, were sequenced by using a genomic DNA sample from the patient. The patient had the homozygous mutation c.115+6T>C (p.Arg10ArgfsX1), which is a GPP-causing mutation that was found in both Chinese and Japanese cohorts [4–6] (Fig. 1c).

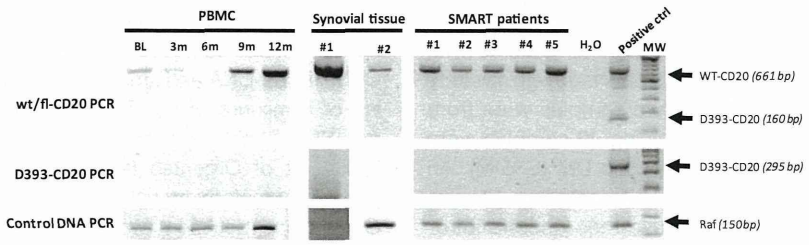
We previously reported *IL36RN* c.115+6T>C as a founder mutation (haplotype; ACTACACC) in a Japanese GPP cohort [4]. Later, in Japanese GPP and IH cases [2,7], we found another haplotype (ACCGAGCC) of c.115+6T>C and herein report the haplotype for the first time. The analysis method for the haplotype of *IL36RN* was described previously [4]. The present Chinese patient also had the haplotype (ACCGAGCC). Thus, the haplotype seems to be shared by the Chinese and Japanese populations.

The prevalence of the *IL36RN* mutation c.115+6T>C is 0.90% (10/1,114 individuals) in the Japanese population and 4.1% (15/365 individuals) in the Chinese population [6,8]. However, the prevalence of the *IL36RN* mutation c.115+6T>C of the specific haplotype (haplotype: ACCGAGCC) in both populations is not known. The *IL36RN* mutation c.115+6T>C (haplotype: ACTACACC) has not been reported in the Chinese population. However, independent from the haplotype, it might be an IH-causing mutation in the Chinese population.

Several twin or sibling cases of IH have been reported [9,10]. Therefore, IH has been thought to be a genetic disease, although the genetic background had been unknown. To date, we have sequenced *IL36RN* in 3 IH cases, including the present case, and found that all of the 3 cases had *IL36RN* mutations [2]. Only a small number of IH patients have been studied genetically, including the present case; thus, further studies of a large number of IH patients are needed in the future.

Abbreviations: GPP, Generalized pustular psoriasis; IH, Impetigo herpetiformis; IL-36, RNInterleukin-36 receptor antagonist; PV, psoriasis vulgaris.

FIG. 1 Alternative CD20 transcript variant expression in PBMCs and synovial tissue from patients with RA



Representative qualitative RT-PCR analysis of *wt/fl cd20* and *d393-cd20* transcripts performed on cDNA from PBMCs of a RA patient, sampled at baseline (BL) and 3, 6, 9 and 12 months after RTX treatment, from two synovial tissues sampled during arthroplasty and from five PBMC samples representative of the SMART cohort (non-responder RTX-treated patients). *fl/wt-cd20* PCR allowed amplification of both *fl/wt-cd20* and *d393-cd20* transcripts, whereas *d393-cd20* PCR amplified specifically the *d393-cd20* transcripts, using a primer spanning the splicing junction. H_2O was used as negative control and cDNA from a B-cell line (positive ctrl) was used as positive control. MW: 100 bp molecular marker. PBMC: peripheral blood mononuclear cells.

Rheumatology key message

- The alternative CD20 transcript is not a marker for resistance to rituximab in RA.

Funding: The French Agence Nationale de la Recherche (Labex LipSTIC, ANR-11-LABX-0021, InflameX, ANR-10-LABX-00) and the Conseil Régional de Franche-Comté ('soutien au LabEX LipSTiC' 2014).

Disclosure statement: The authors have declared no conflicts of interest.

Clémentine Gamonet[1,2,3], Marina Deschamps[1,2,3], Sandrine Marion[4], Georges Herbein[5,6], Gilles Chiocchia[4], Isabelle Auger[7], Philippe Saas[1,2,3,8,9], Christophe Ferrand[1,2,3,8] and Eric Toussirot[6,9,10,11,12]

[1]*INSERM*, [2]*Etablissement Français du Sang Bourgogne Franche Comté*, [3]*Université de Franche-Comté, UMR1098, Besançon*, [4]*INSERM U987, University Versailles Saint Quentin, Simone Veil Department of Health Science, Chronic Inflammation and Immune System, LabEX InflameX, Montigny le Bretonneux*, [5]*CHRU Besançon, Virologie*, [6]*Université de Franche Comté, UPRES EA 4266 Agents Pathogènes et Inflammation, Besançon*, [7]*INSERM, UMR1097, Université Aux Marseille, Marseille*, [8]*Etablissement Français du Sang, Plateforme de Biomonitoring, LabEX LipSTIC*, [9]*INSERM, CIC 1431, Centre investigation Clinique Biothérapie*, [10]*Fédération Hospitalo-Universitaire INCREASE, CHRU*, [11]*CHRU Besançon, Rhumatologie and* [12]*Université de Franche Comté, Département Universitaire de Thérapeutique, Besançon, France*

Revised version accepted 24 April 2015

Correspondence to: Eric Toussirot, Clinical Investigation Center Biotherapy, INSERM, CIC-1431, University Hospital of Besançon, 25000 Besançon, France.
E-mail: etoussirot@chu-besancon.fr

References

1 Benucci M, Manfredi M, Puttini PS, Atzeni F. Predictive factors of response to rituximab therapy in rheumatoid arthritis: what do we know today? Autoimmun Rev 2010;9:801–3.

2 Henry C, Deschamps M, Rohrlich PS *et al*. Identification of an alternative CD20 transcript variant in B-cell malignancies coding for a novel protein associated to rituximab resistance. Blood 2010;115:2420–9.

3 Gamonet C, Ferrand C, Colliou N *et al*. Lack of expression of an alternative CD20 transcript variant in circulating B cells from patients with pemphigus. Exp Dermatol 2014;23:66–7.

4 Mariette X, Rouanet S, Sibilia J *et al*. Evaluation of low-dose rituximab for the retreatment of patients with active rheumatoid arthritis: a non-inferiority randomised controlled trial. Ann Rheum Dis 2014;73:1508–14.

Rheumatology 2015;54:1745–1747
doi:10.1093/rheumatology/kev247
Advance Access publication 9 July 2015

High incidence of cancer in anti-small ubiquitin-like modifier activating enzyme antibody-positive dermatomyositis

SIR, The idiopathic inflammatory myopathies (IIMs) are a group of systemic autoimmune diseases that include PM and DM [1]. Several myositis-specific autoantibodies, which have been regarded as mutually exclusive, are associated with certain clinical forms of IIM.

Since autoantibodies to small ubiquitin-like modifier activating enzyme (SAE) in patients with DM were described [2, 3], a few studies on anti-SAE antibodies in DM have been published from Italy [4], Japan [5] and Hungary [6]. We analysed serum samples from 110 DM patients and 2 were found to be anti-SAE positive [7]. The frequency of anti-SAE antibodies in DM overall was 1.5–5.7%. Nearly all patients with anti-SAE antibodies had skin and muscle symptoms, and most of them had skin disease before the muscle disease; however, the clinical features of the patients with anti-SAE antibodies are

not conclusive. We aimed to establish a quantitative assay for measuring anti-SAE antibodies and to clarify the clinical features of DM patients with these antibodies.

We screened 134 consecutive Japanese patients with DM (12 children, 122 adults) followed at Nagoya University Hospital, Nagoya, Japan. The serum samples were from 85 patients with DM and the remaining 49 samples were from patients with clinically amyopathic DM (CADM). An additional 16 adult patients with DM, including 11 with CADM, were also screened because their doctors introduced them for investigation of DM-marker autoantibodies. Of these 150 patients (male:female ratio 41:109), 67 patients were complicated with interstitial lung disease as diagnosed by chest radiograph or chest CT scan and 22 patients were diagnosed with cancer-associated DM. The definitions of DM, CADM and cancer-associated DM are as defined in our previous study [7]. This study was approved by the ethics committee of Nagoya University. All the patients and healthy individuals provided written informed consent according to the Declaration of Helsinki.

The full-length cDNA clones of SAE1 and SAE2 were purchased from Thermo Scientific Open Biosystems (Waltham, MA, USA). Biotinylated recombinant proteins were produced from the cDNA, using the SP6 Quick Coupled Transcription/Translation System (Promega, Madison, WI, USA). Antibodies against SAE1 and SAE2 were tested by antigen-capture ELISA according to our published protocols [8]. Cut-off values were determined as the mean (+ 5 S.D.) of the units obtained from 36 control serum samples from healthy individuals. Anti-MDA5, anti-Mi-2, anti-NXP-2 and anti-TIF1γ antibodies were also measured.

Serum samples that were positive for anti-SAE by ELISA were analysed with IIF using the Fluoro HEPANA Test (MBL, Nagoya, Japan). The samples were also screened by ELISA kits for antibodies against SS-A/Ro60, SS-B, U1-RNP, Sm, CENP-B, ds-DNA and aminoacyl tRNA synthetases consisting of a mixture of EJ, Jo-1, KS, PL-7 and PL-12 (MBL). Anti-SS-A/Ro52 antibodies were measured by using the ELISA kit of Orgentec (Mainz, Germany). Fisher's exact probability tests were used for comparison of frequencies. Correlations between two parameters were analysed by Spearman's correlation coefficients.

In the first cohort, consisting of 134 serum samples from consecutive patients, 4 (3.0%) patients were positive for both anti-SAE1 and anti-SAE2 antibodies (supplementary Fig. S1A, available at *Rheumatology* Online). Serum samples from two patients had been shown to be positive for anti-SAE antibodies by immunoprecipitation and western blotting [7]. In an additional cohort of 16 patients, 3 had anti-SAE1 antibodies, and 2 of these also had anti-SAE2 antibodies. Anti-SAE1 and anti-SAE2 titres had mutually significant positive correlations ($R = 0.807$, $P < 0.0284$). After the initial screening by ELISA, we investigated anti-SAE antibodies in the serum of five new anti-SAE-positive candidate patients for their ability to immunoprecipitate biotinylated recombinant SAE1 and SAE2. All of the candidates immunoprecipitated recombinant SAE1 and SAE2 (supplementary Fig. S1B, available at *Rheumatology* Online). According to these results, we concluded that we found five new serum samples that were positive for anti-SAE antibodies. All seven of the anti-SAE antibody-positive serum samples exhibited nuclear speckled patterns by IIF analysis (Table 1). Surprisingly, ELISA and immunoprecipitation using

TABLE 1 Clinical characteristics of DM patients with anti-small ubiquitin-like modifier activating enzyme antibody

	Patient 1	Patient 2	Patient 3	Patient 4	Patient 5	Patient 6	Patient 7	Total ($n = 7$), %
Age at onset, years	57	70	65	55	65	77	66	Mean, 65
Sex	Female	Male	Female	Male	Male	Male	Female	4 male:3 female
Heliotrope	+	+	–	+	–	–	–	43
Gottron's sign	+	+	+	+	+	+	+	100
Periungual lesions	+	–	+	+	+	+	NA	83
Mechanic's hands	–	–	–	+	+	+	–	43
V-neck sign	+	–	+	+	–	+	+	71
Shawl sign	+	–	+	+	–	–	+	43
Dysphagia	–	–	–	–	+	+	+	43
Muscle weakness	+	+	+	+	+	–	+	86
Creatine kinase, IU/l	542	429	662	6133	311	5187	1084	Elevated, 100
Interstitial lung disease	+	–	–	+	+	+	–	57
Arthritis	–	–	–	–	NA	–	–	0
Malignancy[a]	–	Rectum	Uterus	–	Oesophagus	Colon	–	57
RP	–	–	–	–	NA	–	–	0
Calcinosis	–	–	–	–	NA	NA	–	0
Other features	PH	–	–	Dysphonia	–	–	–	
Presentation (months)	S (2)	S/M	S (2)	S/M	S (7)	S/M	S (2)	S (mean, 1.9)
Other autoantibodies	Ro52	–	NXP-2, Ro60	Ro60	–	–	–	Ro60 (2), Ro52 (1), NXP-2 (1)

[a]Malignancy associated with DM was defined as that occurring within 3 years of the DM diagnosis. NA: not available; PH: pulmonary hypertension; S: skin disease presented first; S/M: skin and muscle disease presented together.

recombinant NXP-2 protein clarified that one patient also had anti-NXP-2 antibody (data not shown).

Seven anti-SAE-positive patients were diagnosed with adult DM, and all had internal involvement, such as interstitial lung disease, cancer and/or dysphagia, except for patient 7. The frequency of cancer in the anti-SAE-positive patients was significantly higher than in the anti-SAE-negative patients (4/7 *vs* 18/143, $P < 0.0093$). Since anti-NXP-2 antibodies in adult patients with DM are associated with cancer [1], we recalculated the association between anti-SAE antibodies and cancer when the patient with both anti-SAE and anti-NXP-2 antibodies was excluded. The significant association was still confirmed (3/6 *vs* 18/143, $P < 0.0369$).

Previous studies reported the frequency of cancer in anti-SAE-positive patients as 14–25% [3–6]. Interestingly, the cumulative results including our data showed that there were significantly more male patients in the anti-SAE-positive adult cancer-associated myositis group than in the myositis group without cancer (supplementary Table S1, available at *Rheumatology* Online). Multivariate analysis using a large cohort will be needed to clarify whether anti-SAE antibodies independently contribute to the specific clinical characteristics.

Rheumatology key message

- Risk of malignancy should be considered in anti-small ubiquitin-like modifier activating enzyme antibody-positive adult DM patients.

Funding: This work was supported by grants-in-aid for research from the Ministry of Education, Culture, Sports, Science and Technology of Japan (26461656) and for intractable diseases from the Ministry of Health, Labour and Welfare of Japan.

Disclosure statement: The authors have declared no conflicts of interest.

Yoshinao Muro[1], Kazumitsu Sugiura[1], Mizuho Nara[2], Izumi Sakamoto[3], Noriyuki Suzuki[4] and Masashi Akiyama[1]

[1]*Department of Dermatology, Nagoya University Graduate School of Medicine, Nagoya,* [2]*Department of Hematology, Nephrology, and Rheumatology, Akita University Graduate School of Medicine, Akita,* [3]*Department of Nephrology, Nagoya Memorial Hospital, Nagoya and* [4]*Department of Dermatology, Toyohashi Municipal Hospital, Toyohashi, Japan*
Revised version accepted 29 May 2015
Correspondence to: Yoshinao Muro, Division of Connective Tissue Disease and Autoimmunity, Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan. E-mail: ymuro@med.nagoya-u.ac.jp

Supplementary data

Supplementary data are available at *Rheumatology* Online.

References

1 Tansley SL, Betteridge ZE, McHugh NJ. The diagnostic utility of autoantibodies in adult and juvenile myositis. Curr Opin Rheumatol 2013;25:772–7.

2 Betteridge Z, Gunawardena H, North J, McHugh N. Identification of a novel autoantibody directed against small ubiquitin-like modifier activating enzyme in dermatomyositis. Arthritis Rheum 2007;56:3132–7.

3 Betteridge ZE, Gunawardena H, Chinoy H *et al*. Clinical and human leucocyte antigen class II haplotype associations of autoantibodies to small ubiquitin-like modifier enzyme, a dermatomyositis-specific autoantigen target in UK Caucasian adult-onset myositis. Ann Rheum Dis 2009;68:1621–5.

4 Tarricone E, Ghirardello A, Rampudda M *et al*. Anti-SAE antibodies in autoimmune myositis: identification by unlabelled protein immunoprecipitation in an Italian patient cohort. J Immunol Methods 2012;384:128–34.

5 Fujimoto M, Matsushita T, Hamaguchi Y *et al*. Autoantibodies to small ubiquitin-like modifier activating enzymes in Japanese patients with dermatomyositis: comparison with a UK Caucasian cohort. Ann Rheum Dis 2013;72:151–3.

6 Bodoki L, Nagy-Vincze M, Griger Z *et al*. Four dermatomyositis-specific autoantibodies-anti-TIF1γ, anti-NXP2, anti-SAE and anti-MDA5-in adult and juvenile patients with idiopathic inflammatory myopathies in a Hungarian cohort. Autoimmun Rev 2014;13:1211–9.

7 Muro Y, Sugiura K, Akiyama M. Low prevalence of anti-small ubiquitin-like modifier activating enzyme antibodies in dermatomyositis patients. Autoimmunity 2013;46:279–84.

8 Muro Y, Sugiura K, Akiyama M. A new ELISA for dermatomyositis autoantibodies: rapid introduction of autoantigen cDNA to recombinant assays for autoantibody measurement. Clin Dev Immunol 2013;2013:856815.

Rheumatology 2015;54:1747–1749
doi:10.1093/rheumatology/kev221
Advance Access publication 11 June 2015

Tocilizumab in the treatment of a polyostotic variant of fibrous dysplasia of bone

Sir, Fibrous dysplasia of bone (FDB) is a benign disease leading to the slow replacement of normal bone by fibrous tissue, without osteoblastic rimming [1]. Three-quarters of FDB cases are monostotic and occur mainly in craniofacial bones, ribs, femurs and tibias. Polyostotic forms involve, in decreasing order of frequency, femurs, tibias, skull and facial bones, humerus and cervical spine. When associated with café-au-lait macules and hyperfunctioning endocrinopathies, the disease is identified as McCune–Albright syndrome [2]. Bone homeostasis is regulated by the balance between osteoblasts, which build up bone, and osteoclasts, which degrade bone. Pathophysiology of FDB is secondary to an activating mutation in the gene *GNAS* that leads to undifferentiated bone marrow stromal cell

Topical minoxidil improves congenital hypotrichosis caused by *LIPH* mutations

DOI: 10.1111/bjd.13790

Dear Editor, Mutations in *LIPH* are one cause of autosomal recessive woolly hair/hypotrichosis (ARWH).[1] *LIPH* mutations are not uncommon and are found all over the world. In this report, we present four patients with ARWH with *LIPH* mutations who showed hair growth after application of topical minoxidil.

Four nonconsanguineous Japanese patients with ARWH who used 1% or 5% topical minoxidil were observed and followed up. Topical minoxidil is used for androgenic alopecia and is available over the counter in Japan. The present patients used it on their own initiative. Direct sequencing of exon 6 of the *LIPH* gene revealed homozygous c.736T>A (p.Cys246Ser) mutations in patients A and B. The homozygous c.742C>A (p.His248Asn) mutation was found in patient C, and compound heterozygous c.736T>A and c.742C>A mutations were

Table 1 Mutations in the *LIPH* gene in four patients and application of topical minoxidil

Patient	Age (years)	Sex	*LIPH* mutation	Concentration of topical minoxidil (%)	Application period until observation (months)
A	3	Male	c.736 T>A homozygous	1 and 5	31
B	7	Female	c.736 T>A homozygous	1	17
C	70	Female	c.742C>A homozygous	1	6
D	5	Female	c.736 T>A, c.742C>A compound heterozygous	1	55

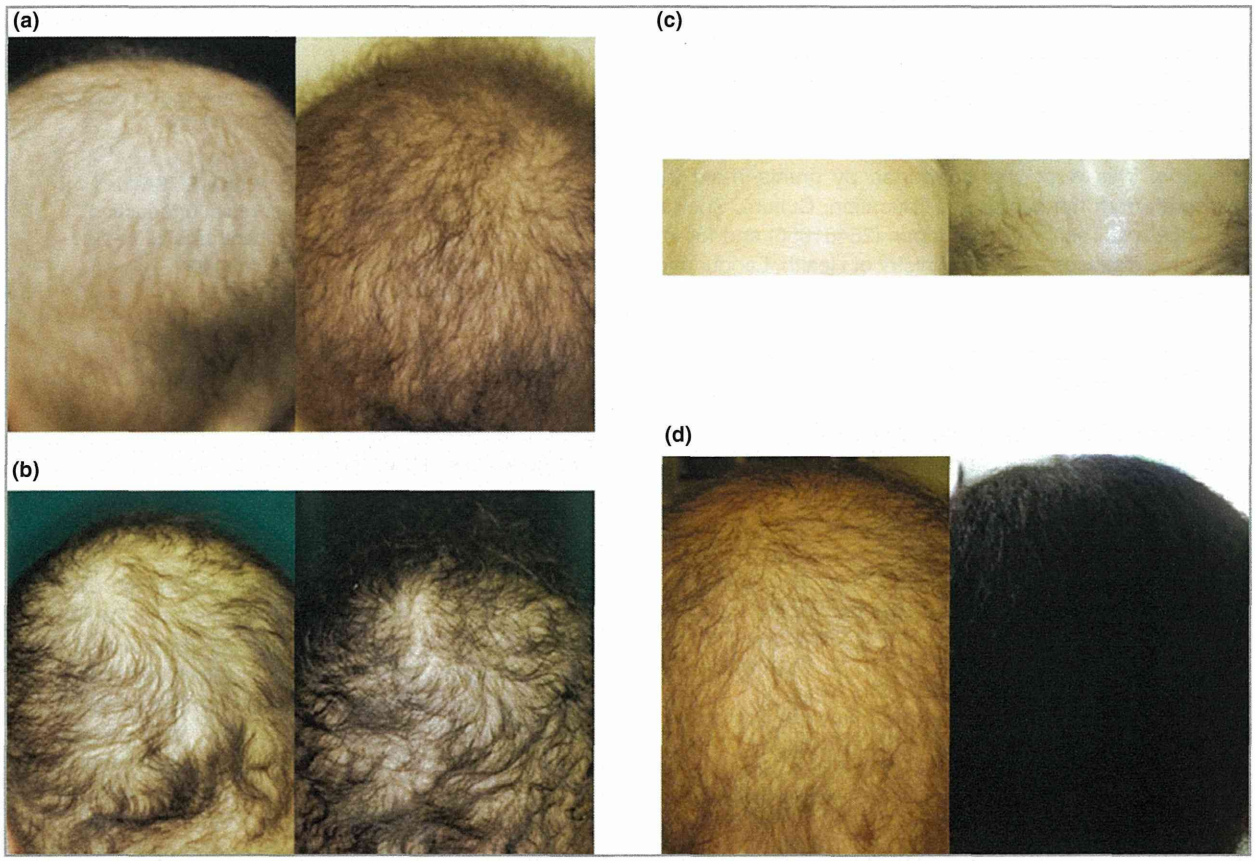


Fig 1. Clinical features of the present patients before and after topical minoxidil application. (a) Patient A, (b) Patient B, (c) Patient C and (d) Patient D. Left, photos before topical minoxidil application; right, photos after topical minoxidil application. Patient A after 2·5 years' treatment (1% topical minoxidil in the first year, 5% topical minoxidil during the second year); Patient B after 1·5 years' treatment (1% topical minoxidil); Patient C after 6 months of treatment (1% topical minoxidil application); Patient D after 4·5 years of topical minoxidil application.

found in patient D (Table 1). Patients A and C were included in previous reports, but their use of topical minoxidil was unknown at the time.[2,3] The mutation search was approved by the Medical Ethics Committee of Nagoya University Graduate School of Medicine and was conducted according to the Declaration of Helsinki principles. The patients each provided written informed consent. The patients had used 1% or 5% topical minoxidil of their own will for 6 months to 3 years. Amazingly, the hair of all these patients grew after use of topical minoxidil, and all patients felt that they benefited from the treatment (Table 1, Fig. 1). In particular, patient C had little hair growth from childhood to the age of 70, but after topical minoxidil application, her hair grew visibly for the first time in her life. No patient claimed adverse effects, such as hypertrichosis of other body parts or cardiovascular problems, during the periods of use.

Mutations in *LIPH* are one cause of ARWH. *LIPH* encodes a membrane-associated phosphatidic acid-preferring phospholipase $A_1\alpha$, which produces lysophosphatidic acid from phosphatidic acid and plays a crucial role in hair growth in humans.[1]

The two missense mutations, c.736T>A and c.742C>A, are considered the prevalent founder mutations in Japanese ARWH patients, and the combined carrier rate of the mutations is about 2·1% in the Japanese population.[3] Hence, as many as 10 000 ARWH patients are estimated to exist in Japan. Unfortunately, no effective treatment has yet been established, and the hypotrichosis significantly decreases quality of life of the patients for almost their entire lifetime.

As well as the four patients with ARWH and *LIPH* mutations described here, we recently reported another case of ARWH that improved with topical minoxidil.[2] The mechanism by which topical minoxidil affects hair growth is not fully understood, although it is thought to have an effect on cell growth and duration of the anagen hair growth phase and to enlarge miniaturized follicles. Topical minoxidil has generally been used for androgenic alopecia but has also been reported to be effective for other hair loss, including congenital hypotrichosis, such as ectodermal dysplasia.[4] The present cases suggest minoxidil could improve congenital hypotrichosis due to LIPH mutations, and it may also potentially improve congenital hypotrichosis due to mutations in other causative genes.

The most frequent adverse effect of minoxidil is hypertrichosis, usually localized to the head. Additionally, adverse cardiovascular effects, such as sinus tachycardia, palpitation and dizziness, have been reported with 2% topical minoxidil use in three patients aged from 10 to 14 years, all of whom recovered from these adverse effects after discontinuance of minoxidil.[5] Although these adverse effects are rare, topical minoxidil must be used carefully, especially in children.

In conclusion, combined with a previous report, improvement of hypotrichosis was observed after topical minoxidil application in five cases of ARWH due to LIPH mutations. These cases suggest that topical minoxidil could be useful for treating congenital hypotrichosis caused by *LIPH* mutations.

Acknowledgments

The authors thank Ms Haruka Ozeki and Ms Yuka Terashita for their technical assistance in analysing *LIPH* mutations.

Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan
K. Tanahashi
K. Sugiura
M. Akiyama
Correspondence: Masashi Akiyama.
E-mail: makiyama@med.nagoya-u.ac.jp

References

1 Pasternack SM, von Kugelgen I, Al Aboud K *et al.* G protein-coupled receptor P2Y5 and its ligand LPA are involved in maintenance of human hair growth. *Nat Genet* 2008; **40**:29–34.
2 Tanahashi K, Sugiura K, Takeichi T *et al.* Prevalent founder mutation c.736T>A of LIPH in autosomal recessive woolly hair of Japanese leads to variable severity of hypotrichosis in adulthood. *J Eur Acad Dermatol Venereol* 2013; **27**:1182–4.
3 Tanahashi K, Sugiura K, Kono M *et al.* Highly prevalent LIPH founder mutations causing autosomal recessive woolly hair/hypotrichosis in Japan and the genotype/phenotype correlations. *PLoS One* 2014; **9**:e89261.
4 Lee HE, Chang IK, Im M *et al.* Topical minoxidil treatment for congenital alopecia in hypohidrotic ectodermal dysplasia. *J Am Acad Dermatol* 2013; **68**:e139–40.
5 Georgala S, Befon A, Maniatopoulou E *et al.* Topical use of minoxidil in children and systemic side effects. *Dermatology* 2007; **214**: 101–2.

Funding sources: This study was supported in part by Grant-in-Aid for Challenging Exploratory Research 26670526 (to K.S.) and Grant-in-Aid for Scientific Research (A) 23249058 (to M.A.), both from the Ministry of Education, Culture, Sports, Science, and Technology of Japan.

Conflicts of interest: none declared.

White piedra caused by *Trichosporon inkin*: a report of two cases in a northern climate

DOI: 10.1111/bjd.13824

Dear Editor, We recently diagnosed two patients attending a specialty hair clinic in Boston, with white piedra, a superficial fungal infection caused by several species of *Trichosporon* that affects the terminal hair shaft. This is notable because white piedra is said to be rare in the U.S.A., particularly in northern climates. The causative species and the laboratory techniques used to identify the organism are of interest, as the organism is difficult to culture and taxonomy has recently changed.

Chao-Kai Hsu[b,c]
[b]St. John's Institute of Dermatology, King's College London, Guy's Hospital, Great Maze Pond, London SE1 9RT, UK; [c]Department of Dermatology, National Cheng Kung University Hospital, College of Medicine, National Cheng Kung University, Tainan, Taiwan

Kana Tanahashi[a]
[a]Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan

Hiroyuki Takama[a]
[a]Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan

Michael A. Simpson[d]
[d]Division of Genetics and Molecular Medicine, King's College London, Guy's Hospital, Great Maze Pond, London SE1 9RT, UK

John A. McGrath[b]
[b]St. John's Institute of Dermatology, King's College London, Guy's Hospital, Great Maze Pond, London SE1 9RT, UK

Masashi Akiyama[a,*]
[a]Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan

*Corresponding author. Fax: +81 52 744 2318
E-mail address: makiyama@med.nagoya-u.ac.jp (M. Akiyama).

Received 28 May 2015
Received in revised form 24 June 2015
Accepted 1 July 2015

http://dx.doi.org/10.1016/j.jdermsci.2015.07.001

Letter to the Editor

Impetigo herpetiformis with *IL36RN* mutations in a Chinese patient: A founder haplotype of c.115+6T>C in East Asia

Keywords:
Founder haplotype
Generalized pustular psoriasis
IL36RN
Impetigo herpetiformis
Interleukin-36 receptor antagonist

Impetigo herpetiformis (IH) is a rare pustular dermatosis of pregnancy [1]. Most patients with IH do not have personal and family histories of psoriasis. Early diagnosis is essential, as IH occasionally leads to maternal or foetal death. Despite the clinical importance of IH, its aetiology has not been clarified sufficiently. Recently, we reported two Japanese cases of IH with homozygous and heterozygous mutations of *IL36RN*, which encodes the interleukin-36 receptor antagonist (IL-36RN) [2]. However, the incidence of IH cases with *IL36RN* mutations is unknown. To our knowledge, no subsequent case of IH with or without an *IL36RN* mutation has been reported thus far.

After a long-standing controversy over whether IH is an independent disease entity from generalized pustular psoriasis (GPP), today there is a tentative consensus that IH is GPP occurring during pregnancy [3]. We reported that most GPP cases that are not accompanied by psoriasis vulgaris (PV; GPP alone) are caused by *IL36RN* mutations, although only a small number of cases with GPP preceding or accompanied by PV were found to have *IL36RN* mutations [4].

Here, we report a case of IH in a Chinese patient with a homozygous *IL36RN* mutation c.115+6T>C, the most frequent GPP-causing mutation in the Chinese population. We also found a novel haplotype of *IL36RN* c.115+6T>C, which is a probable founder haplotype, both in the present patient and in 2 Japanese families.

The patient was a 25-year-old Chinese woman who was admitted to our hospital for pustular lesions after her first normal vaginal delivery (Fig. 1a). She had neither a family history of GPP and IH nor consanguinity in her family. She had no history of GPP. Her pustular lesions began to develop at the 29th week of her first pregnancy, and she had been treated in a maternity hospital. Oral betamethasone of 3 mg/day was administrated, although the eruptions persisted. The skin biopsy of a specimen from a pustular eruption on the trunk revealed a spongiform pustule of Kogoj in the epidermis, consistent with IH (Fig. 1b). She had erythema with pustules all over her body and fever of a body temperature higher than 38 °C. Blood examinations revealed white blood cell counts of 31,590/μL and C-reactive protein concentrations of 2.45 mg/dL (reference range: <0.3 mg/dL). Bacterial culture of the pustules yielded negative results. Thus, she was diagnosed as having IH.

After ethical approval, informed consent was obtained in compliance with the guidelines of the Declaration of Helsinki. The entire coding regions of *IL36RN*, including the exon/intron boundaries, were sequenced by using a genomic DNA sample from the patient. The patient had the homozygous mutation c.115+6T>C (p.Arg10ArgfsX1), which is a GPP-causing mutation that was found in both Chinese and Japanese cohorts [4–6] (Fig. 1c).

We previously reported *IL36RN* c.115+6T>C as a founder mutation (haplotype; ACTACACC) in a Japanese GPP cohort [4]. Later, in Japanese GPP and IH cases [2,7], we found another haplotype (ACCGAGCC) of c.115+6T>C and herein report the haplotype for the first time. The analysis method for the haplotype of *IL36RN* was described previously [4]. The present Chinese patient also had the haplotype (ACCGAGCC). Thus, the haplotype seems to be shared by the Chinese and Japanese populations.

The prevalence of the *IL36RN* mutation c.115+6T>C is 0.90% (10/1,114 individuals) in the Japanese population and 4.1% (15/365 individuals) in the Chinese population [6,8]. However, the prevalence of the *IL36RN* mutation c.115+6T>C of the specific haplotype (haplotype: ACCGAGCC) in both populations is not known. The *IL36RN* mutation c.115+6T>C (haplotype: ACTACACC) has not been reported in the Chinese population. However, independent from the haplotype, it might be an IH-causing mutation in the Chinese population.

Several twin or sibling cases of IH have been reported [9,10]. Therefore, IH has been thought to be a genetic disease, although the genetic background had been unknown. To date, we have sequenced *IL36RN* in 3 IH cases, including the present case, and found that all of the 3 cases had *IL36RN* mutations [2]. Only a small number of IH patients have been studied genetically, including the present case; thus, further studies of a large number of IH patients are needed in the future.

Abbreviations: GPP, Generalized pustular psoriasis; IH, Impetigo herpetiformis; IL-36, RNInterleukin-36 receptor antagonist; PV, psoriasis vulgaris.

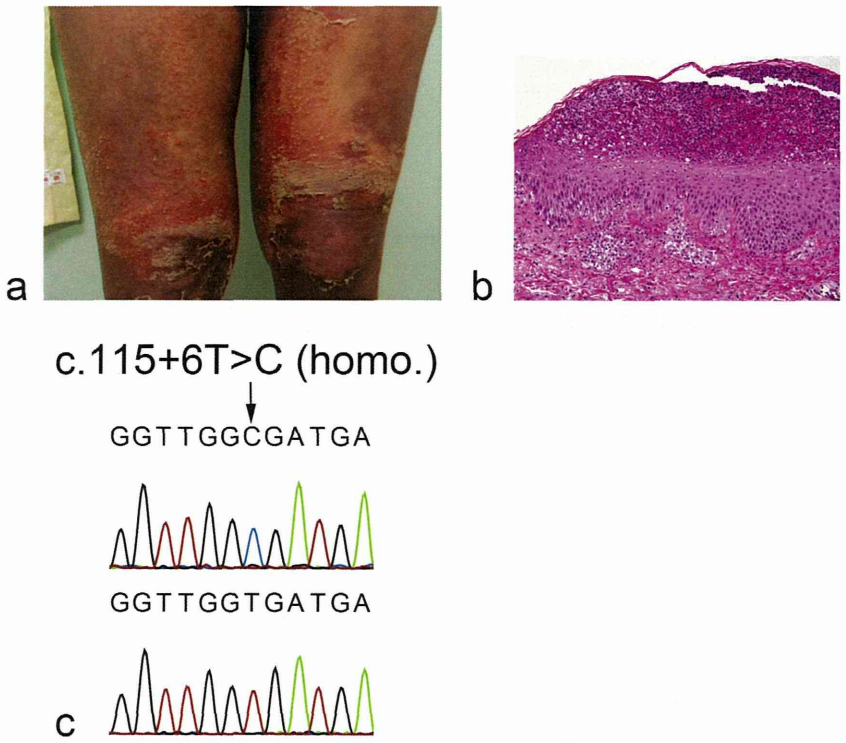


Fig. 1. Clinical and histopathologic features, and mutation analysis of *IL36RN* in the patient
The clinical features of the present case (a). Pustules on the background erythema are seen on the thighs. The pathologic features of the pustules (b). Spongiosis of Kogoj and acanthosis can be seen in the epidermis of the pustular erythematous lesions on the trunk (original magnification: × 200). Direct sequencing reveals the homozygous mutation c.115+6T>C in the present case (c).

In conclusion, the present case further support that IH and GPP, especially GPP alone, are identical diseases caused by *IL36RN* mutations.

Conflicts of interests

The authors have no conflicts of interests to declare.

Acknowledgements

The authors thank Ms. Haruka Ozeki and Ms. Yuka Terashita for their technical help in analysing *IL36RN* mutations. This study was supported in part by a Grant-in-Aid for Scientific Research (B) to K.S. (15H04886), a Grant-in-Aid for Challenging Exploratory Research to K.S. (15K15414), a Grant-in-Aid for Scientific Research (B) to M.A. (15H04887), and a Grant-in-Aid for Challenging Exploratory Research to M.A. (15K15415), from the Ministry of Education, Culture, Sports, Science, and Technology of Japan, and by the grant H26-itaku (nan)-ippan-027 to K.S. from the Ministry of Health, Labour and Welfare (Research on Measures for Intractable Disease), Japan.

Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at http://dx.doi.org/10.1016/j.jdermsci.2015.06.003.

References

[1] G.C. Sauer, B.J. Geha, Impetigo herpetiformis. Report of a case treated with corticosteroid-review of the literature, Arch. Dermatol. 83 (1961) 119–126.
[2] K. Sugiura, N. Oiso, S. Iinuma, H. Matsuda, M. Minami-Hori, A. Ishida-Yamamoto, et al., IL36RN mutations underlie impetigo herpetiformis, J. Invest. Dermatol. 134 (2014) 2472–2474.
[3] A. Robinson, A.S. Van Voorhees, S. Hsu, N.J. Korman, M.G. Lebwohl, B.F. Bebo Jr., et al., Treatment of pustular psoriasis: from the Medical Board of the National Psoriasis Foundation, J. Am. Acad. Dermatol. 67 (2012) 279–288.
[4] K. Sugiura, A. Takemoto, M. Yamaguchi, H. Takahashi, Y. Shoda, T. Mitsuma, et al., The majority of generalized pustular psoriasis without psoriasis vulgaris is caused by deficiency of interleukin-36 receptor antagonist, J. Invest. Dermatol. 133 (2013) 2514–2521.
[5] K. Sugiura, The genetic background of generalized pustular psoriasis: IL36RN mutations and CARD14 gain-of-function variants, J. Dermatol. Sci. 74 (2014) 187–192.
[6] M. Li, J. Han, Z. Lu, H. Li, K. Zhu, R. Cheng, et al., Prevalent and rare mutations in IL-36RN gene in Chinese patients with generalized pustular psoriasis and psoriasis vulgaris, J. Invest. Dermatol. 133 (2013) 2637–2639.
[7] K. Sugiura, K. Endo, T. Akasaka, M. Akiyama, Successful treatment with infliximab of sibling cases with generalized pustular psoriasis caused by deficiency of interleukin -36 receptor antagonist, J. Eur. Acad. Dermatol. Venereol. (2014), http://dx.doi.org/10.1111/jdv.12590.
[8] M. Hayashi, T. Nakayama, T. Hirota, H. Saeki, Y. Nobeyama, T. Ito, et al., Novel IL36RN gene mutation revealed by analysis of 8 Japanese patients with generalized pustular psoriasis, J. Dermatol. Sci. 76 (2014) 267–269.
[9] J. Tada, S. Fukushiro, Y. Fujiwara, Y. Akagi, H. Kodama, N. Nohara, Two sisters with impetigo herpetiformis, Clin. Exp. Dermatol. 14 (1989) 82–84.
[10] K. Vicdan, Z. Gokay, T. Var, N. Danisman, O. Gokmen, Twin sisters with impetigo herpetiformis, Eur. J. Obstet. Gynecol. Reprod. Biol. 63 (1995) 195–196.

Kazumitsu Sugiura[a,*], Ayaka Nakasuka[b], Hiroaki Kono[b], Michihiro Kono[a], Masashi Akiyama[a]
[a]*Department of Dermatology, Nagoya University Graduate School of Medicine, 65 Tsurumai-cho, Showa-ku, Nagoya 466-8550, Japan*;
[b]*Department of Dermatology, Kochi Health Science Center, 2125-1 Ike, Kochi 781-8555, Japan*

*Corresponding author. Tel.: +81 52 744 2314;
fax: +81 52 744 2318
E-mail address: kazusugi@med.nagoya-u.ac.jp (K. Sugiura).

Received 4 March 2015
Received in revised form 3 June 2015
Accepted 5 June 2015

http://dx.doi.org/10.1016/j.jdermsci.2015.06.003

Research letter

Dowling–Degos disease with mutations in *POFUT1* is clinicopathologically distinct from reticulate acropigmentation of Kitamura

DOI: 10.1111/bjd.13702

Dear Editor, Dowling–Degos disease (DDD) is a rare autosomal dominant genetic pigmentary disorder characterized by dot-like or reticulate, slightly depressed, sharply demarcated brown macules particularly affecting the flexures and other major skinfolds (Fig. 1a).[1] There has long been controversy over whether DDD and reticulate acropigmentation of Kitamura (RAK; MIM #615537), which has similar skin manifestations but affects mainly the dorsa of the hands and the feet, are distinct clinical entities or variants of the same disease. Several reports have suggested that RAK and DDD are identical disorders with different spectra.[2,3] The causative gene of DDD was clarified as *KRT5* in 2006,[4] and recently also *POFUT1* (encoding protein *O*-fucosyltransferase 1)[5] and *POGLUT1*[6] were identified. As for RAK, it was shown to be due to mutations in *ADAM10* in 2013.[7]

In this study, to clarify the differences between genetically confirmed DDD and RAK, we performed genetic diagnoses of three DDD pedigrees and one RAK pedigree. We compared the detailed clinical and histological features between patients with DDD and patients with RAK, who were confirmed to have the causative *POFUT1* and *ADAM10* gene mutations, respectively, including previously reported cases.

The mutation search for each gene was performed as previously described.[4–7] Informed consent and blood samples of patients were obtained under protocols approved by the ethics review committee of Nagoya University School of Medicine. In addition, histopathological examinations of biopsy specimens from the skin lesions were performed.

Four patients with DDD from three unrelated families, who had the known heterozygous mutation c.397C>T (p.Arg133X)[8] (family D1) or the novel mutations c.460C>T (p.Gln154X) (family D2) or c.891G>A (p.Trp297X) (family D3) in *POFUT1*, were included in the present study (Figs S1 and S2a–c; see Supporting Information). Two patients with RAK from one family who had the novel heterozygous mutation c.1000G>A (p.Gly334Arg) (family R1) in *ADAM10* were also included in the present study (Fig. S2d,e). In addition, we referred to nine cases of RAK previously reported by our group[7] and two cases of DDD reported by Li *et al.* (Table S1; see Supporting Information).[5]

The histopathological features of the patients with DDD with the *POFUT1* mutations were acanthosis of the epidermis, tight digitiform rete ridges with prominent hyperpigmentation at the tips, pigmentary incontinence and small cornified cysts (Fig. 1e). In contrast, histopathological investigation revealed that the epidermis of the patient with RAK with the *ADAM10* mutation showed pigmentation at the tip of the rete ridges, slight elongation and thinning of the rete ridges, thinning of the epidermis and slight hyperkeratosis without parakeratosis or pigmentary incontinence (Fig. 1f).

Both *POFUT1* and *POGLUT1* are involved in the Notch pathway.[6] Keratinocyte-specific deletion of the *Notch1* gene results in marked epidermal hyperplasia.[9] These investigational results may support the idea of histopathological differences between DDD and RAK. Thus, DDD skin lesions show acanthosis with tight digitiform rete ridges, although *Adam10*-deficient mice show thinning of the spinous layers of the epidermis.[10] Regarding melanocytes, although *Adam10*-deficient mice show no alteration of pigmentation, *Adam10*-deficient hairless mice show pigmented macules.[11] Notch signalling may also affect the melanocyte lineage. Genetic ablation of Notch signalling in the mouse results in a dramatic reduction of embryonic melanoblasts and a dilution of initial hair pigmentation.[12,13] However, it is still unknown how these research results relate to the pathogenesis of pigmentation diseases in humans.

The age at onset and the distribution of the skin lesions also differ between DDD and RAK (Table S1). RAK has an earlier age of onset than DDD: the age of onset for DDD ranges from 18 to 56 years, averaging 28·8 ± 13·9 years, whereas the onset age for RAK ranges from 5 to 12 years, averaging 9·2 ± 2·2 years. Among all of the 11 patients with RAK in this study and in our previous report, 10 had the initial skin lesion on the dorsa of the hands. In contrast, four of the five patients with DDD with information available on the primary sites had the primary skin lesions at locations other than the dorsa of the hands. Comedo-like follicular papules were seen only in the patients with DDD (Fig. 1c) and not in any patient with RAK. Hyperpigmentation and papules on the perianal and genital regions were also seen only in patients with DDD (Fig. 1d)[14] and not in RAK.

All of the patients with DDD except D3-1 showed skin manifestations involving the limbs. Interestingly, all of their mutations were truncation mutations around the N-terminal third of POFUT-1, abolishing the C-terminal two-thirds of the amino acid sequence of the protein (Fig. 1g). In contrast, case D3-1 showed a clinically rare genital lesion; histopathologically, the acanthosis and digitiform rete ridges were relatively mild, although the hyperpigmentation at the tips still stood out. The *POFUT1* truncation mutation in case D3-1 results in

