Kikuchi K, Natsuga K, Shinkuma S, Nishie W, Kajita S, Sato H, **Shimizu H**	Subepidermal blistering disease with 3 distinct autoantibodies: Anti-BP230, anti-laminin gamma-1, and anti-laminin-332.	**J Am Acad Dermatol**	65	878-880	2011
Kikuchi K, **Arita K**, Tateishi Y, Onozawa M, **Akiyama M**, **Shimizu H**	Recurrence of hydroxyurea-induced leg ulcer after discontinuation of treatment.	**Acta Derm Venereol**	91	373-374	2011
Kikuchi K, Abe R, Shinkuma S, Hamasaka H, Natsuga K, Hata H, Tateishi Y, Shibata M, Tomita Y, Abe A, Aoyagi A, Mukai M, **Shimizu H**	Spontaneous Remission of Solitary-Type Infantile Myofibromatosis.	**Case Rep Dermatol**	3	181-185	2011
Kempf M, Miyamura Y, Liu PY, Chen AC, Nakamura H, **Shimizu H**, Tabata Y, Kimble RM, McMillan JR	A denatured collagen microfiber scaffold seeded with human fibroblasts and keratinocytes for skin grafting.	**Biomaterials**	32	4782-4792	2011
Fujita Y, Yoshioka N, Abe R, Murata J, Hoshina D, Mae H, **Shimizu H**	Rapid immunochromatographic test for serum granulysin is useful for the prediction of Stevens-Johnson syndrome and toxic epidermal necrolysis.	**J Am Acad Dermatol**	65	65-68	2011
Fujita Y, Abe R, Nishie W, **Shimizu H**	Regenerative medicine for severe congenital skin disorders: restoration of deficient skin component proteins by stem cell therapy.	**Inflammation and Regeneration**	31	282-289	2011
Frew J, Lim SW, Klausseger A, Chow CW, Tran K, Su J, Orchard D, Varigos G, Sawamura D, Nishie W, **Shimizu H**, Murrell DF	Autosomal dominant bullous dermolysis of the newborn associated with a heterozygous missense mutation p.G1673R in type VII collagen.	**Australas J Dermatol**	52	e1-4	2011
Chen AC, McNeilly C, Liu AP, Flaim CJ, Cuttle L, Kendall M, Kimble RM, **Shimizu H**, McMillan JR	Second harmonic generation and multiphoton microscopic detection of collagen without the need for species specific antibodies.	**Burns**	37	1001-1009	2011

Fukamachi S, Mori T, Sakabe J-I, Shiraishi N, Kuroda E, Kobayashi M, Bito T, Kabashima K, Nakamura M, **Tokura Y**	Topical Cholecystokinin Depresses Itch-Associated Scratching Behavior in Mice	**J Invest Dermatol**	131	956-961	2011
Sawada Y, Hino R, Hama K, Ohmori S, Fueki H, Yamada S, Fukamachi S, Tajiri M, Kumo R, Yoshioka M, Nakashima D, Sugita K, Yoshiki R, Shimauchi T, Mori T, Izu K, Kobayashi M, Nakamura M, **Tokura Y**	The type of skin eruptions is an independent prognostic indicator for adult T-cell leukemia/lymphoma.	**Blood**	117	3161-3967	2011
Fukamachi S, Bito T, Shiraishi N, Kobayashi M, Kabashima K, Nakamura M, **Tokura Y**	Modulation of semaphorin 3A expression by calcium concentration and histamine in human keratinocytes and fibroblasts.	**J Dermatol Sci**	62	118-123	2011
Yoshihisa Y, Honda A, Zhao QL, Makino T, Abe R, Matsui K, **Shimizu H**, Miyamoto Y, Kondo T, Shimizu T	Protective effects of platinum nanoparticles against UV-light-induced epidermal inflammation.	**Exp Dermatol**	19	1000-1006	2010
Yanagi T, **Shimizu H**, Shimizu T	Occupational contact dermatitis caused by asparagus.	**Contact Dermatitis**	63	54	2010
Yanagi T, **Akiyama M**, Nishihara H, Ishikawa J, Sakai K, Miyamura Y, Naoe A, Kitahara T, Tanaka S, **Shimizu H**	Self-improvement of keratinocyte differentiation defects during skin maturation in ABCA12-deficient harlequin ichthyosis model mice.	**Am J Pathol**	177	106-118	2010
Wang G, Ujiie H, Shibaki A, Nishie W, Tateishi Y, Kikuchi K, Li Q, McMillan JR, Morioka H, Sawamura D, Nakamura H, **Shimizu H**	Blockade of autoantibody-initiated tissue damage by using recombinant fab antibody fragments against pathogenic autoantigen.	**Am J Pathol**	176	914-925	2010
Uo M, Asakura K, Watanabe E, Lin HY, Yanagi T, **Shimizu H**, Watari F	A study of Zinc Contatined in Yellow and Black Discolored Nails by X-ray Fluorescence and X-ray Absorption Fine Structure Analyses.	**Nano Biomedicine**	2	103-106	2010

Ujiie H, Shibaki A, Nishie W, Sawamura D, Wang G, Tateishi Y, Li Q, Moriuchi R, Qiao H, Nakamura H, **Akiyama M**, **Shimizu H**	A novel active mouse model for bullous pemphigoid targeting humanized pathogenic antigen.	**J Immunol**	184	2166-2174	2010
Ujiie H, Shibaki A, **Akiyama M**, **Shimizu H**	Successful treatment of nail lichen planus with topical tacrolimus.	**Acta Derm Venereol**	90	218-219	2010
Ujiie H, Kodama K, **Akiyama M**, **Shimizu H**	Hereditary benign telangiectasia: two families with punctate telangiectasias surrounded by anemic halos.	**Arch Dermatol**	146	98-99	2010
Ujihara M, Nakajima K, Yamamoto M, Teraishi M, Uchida Y, **Akiyama M**, **Shimizu H**, Sano S	Epidermal triglyceride levels are correlated with severity of ichthyosis in Dorfman-Chanarin syndrome.	**J Dermatol Sci**	57	102-107	2010
Uchida Y, Cho Y, Moradian S, Kim J, Nakajima K, Crumrine D, Park K, Ujihara M, **Akiyama M**, **Shimizu H**, Holleran WM, Sano S, Elias PM	Neutral lipid storage leads to acylceramide deficiency, likely contributing to the pathogenesis of Dorfman-Chanarin syndrome.	**J Invest Dermatol**	130	2497-2499	2010
Tsuruta D, **Akiyama M**, Ishida-Yamamoto A, Imanishi H, Mizuno N, Sowa J, Kobayashi H, Ishii M, Kurokawa I, **Shimizu H**	Three-base deletion mutation c.120_122delGTT in ATP2A2 leads to the unique phenotype of comedonal Darier disease.	**Br J Dermatol**	162	687-689	2010
Shinkuma S, Natsuga K, Nishie W, **Shimizu H**	Epidermolysis bullosa in Japan.	**Dermatol Clin**	28	431-432, xvi	2010
Shinkuma S, **Akiyama M**, Inoue A, Aoki J, Natsuga K, **Nomura T**, **Arita K**, Abe R, Ito K, Nakamura H, Ujiie H, Shibaki A, Suga H, Tsunemi Y, Nishie W, **Shimizu H**	Prevalent LIPH founder mutations lead to loss of P2Y5 activation ability of PA-PLA1alpha in autosomal recessive hypotrichosis.	**Hum Mutat**	31	602-610	2010
Shimizu S, Natsuga K, Shinkuma S, Yasui C, Tsuchiya K, **Shimizu H**	Localized Linear IgA/IgG Bullous Dermatosis.	**Acta Derm Venereol**	90	621-624	2010
Sakai K, **Akiyama M**, Yanagi T, Nampoothiri S, Mampilly T, Sunitha V, **Shimizu H**	An Indian family with Sjogren-Larsson syndrome caused by a novel ALDH3A2 mutation.	**Int J Dermatol**	49	1031-1033	2010

Saito N, Yanagi T, **Akiyama M**, Lin HY, Kasai S, Fujita Y, Yamane N, Inokuma D, Kase S, Ono K, Minakawa H, **Shimizu H**	Pyoderma Gangrenosum of the Eyelid: Report of Two Cases and Review of the Literature.	**Dermatology**	221	211-215	2010
Saito N, Yamane N, Matsumura W, Fujita Y, Inokuma D, Kuroshima S, Hamasaka K, **Shimizu H**	Generalized exacerbation of systemic allergic dermatitis due to zinc patch test and dental treatments.	**Contact Dermatitis**	62	372-373	2010
Oji V, Tadini G, **Akiyama M**, Blanchet Bardon C, Bodemer C, Bourrat E, Coudiere P, Digiovanna JJ, Elias P, Fischer J, Fleckman P, Gina M, Harper J, Hashimoto T, Hausser I, Hennies HC, Hohl D, Hovnanian A, Ishida-Yamamoto A, Jacyk WK, Leachman S, Leigh I, Mazereeuw-Hautier J, Milstone L, Morice-Picard F, Paller AS, Richard G, Schmuth M, **Shimizu H**, Sprecher E, Van Steensel M, Taieb A, Toro JR, Vabres P, Vahlquist A, Williams M, Traupe H	Revised nomenclature and classification of inherited ichthyoses: Results of the First Ichthyosis Consensus Conference in Soreze 2009.	**J Am Acad Dermatol**	63	607-641	2010
Natsuga K, Sawamura D, Goto M, Homma E, Goto-Ohguchi Y, Aoyagi S, **Akiyama M**, Kuroyanagi Y, **Shimizu H**	Response of intractable skin ulcers in recessive dystrophic epidermolysis bullosa patients to an allogeneic cultured dermal substitute.	**Acta Derm Venereol**	90	165-169	2010
Natsuga K, Nishie W, Shinkuma S, Moriuchi R, Shibata M, Nishimura M, Hashimoto T, **Shimizu H**	Circulating IgA and IgE autoantibodies in antilaminin-332 mucous membrane pemphigoid.	**Br J Dermatol**	162	513-517	2010
Natsuga K, Nishie W, Shinkuma S, **Arita K**, Nakamura H, Ohyama M, Osaka H, Kambara T, Hirako Y, **Shimizu H**	Plectin deficiency leads to both muscular dystrophy and pyloric atresia in epidermolysis bullosa simplex.	**Hum Mutat**	31	E1687-1698	2010

Natsuga K, Nishie W, **Arita K**, Shinkuma S, Nakamura H, Kubota S, Imakado S, **Akiyama M**, **Shimizu H**	Complete paternal isodisomy of chromosome 17 in junctional epidermolysis bullosa with pyloric atresia.	**J Invest Dermatol**	130	2671-2674	2010
Natsuga K, Nishie W, **Akiyama M**, Nakamura H, Shinkuma S, McMillan JR, Nagasaki A, Has C, Ouchi T, Ishiko A, Hirako Y, Owaribe K, Sawamura D, Bruckner-Tuderman L, **Shimizu H**	Plectin expression patterns determine two distinct subtypes of epidermolysis bullosa simplex.	**Hum Mutat**	31	308-316	2010
Mitsutake S, Suzuki C, **Akiyama M**, Tsuji K, Yanagi T, **Shimizu H**, Igarashi Y	ABCA12 dysfunction causes a disorder in glucosylceramide accumulation during keratinocyte differentiation.	**J Dermatol Sci**	60	128-129	2010
Li Q, Ujiie H, Shibaki A, Wang G, Moriuchi R, Qiao HJ, Morioka H, Shinkuma S, Natsuga K, Long HA, Nishie W, **Shimizu H**	Human IgG1 Monoclonal Antibody against Human Collagen 17 Noncollagenous 16A Domain Induces Blisters via Complement Activation in Experimental Bullous Pemphigoid Model.	**J Immunol**	185	7746-7755	2010
Inokuma D, Shibaki A, **Shimizu H**	Two cases of cutaneous sporotrichosis in continental/microthermal climate zone: global warming alert?	**Clin Exp Dermatol**	35	668-669	2010
Iitani MM, Abe R, Yanagi T, Hamasaka A, Tateishi Y, Abe Y, Ito M, Kondo T, Kubota K, **Shimizu H**	Aleukemic leukemia cutis with extensive bone involvement.	**J Am Acad Dermatol**	63	539-541	2010
Hata H, Aoyagi S, Iitani MM, Homma E, **Shimizu H**	Squamous Cell Carcinoma in a Chronic Genital Ulcer in Behcet's Disease	**Acta Derm Venereol**	90	540-541	2010
Hamasaka A, Yoshioka N, Abe R, Kishino S, Umezawa K, Ozaki M, Todo S, **Shimizu H**:	Topical application of dehydroxymethylepoxyquinomicin improves allergic inflammation via NF-kappaB inhibition.	**J Allergy Clin Immunol**	126	400-403	2010

Fujita Y, Abe R, Inokuma D, Sasaki M, Hoshina D, Natsuga K, Nishie W, McMillan JR, Nakamura H, Shimizu T, **Akiyama M**, Sawamura D, **Shimizu H**	Bone marrow transplantation restores epidermal basement membrane protein expression and rescues epidermolysis bullosa model mice.	**Proc Natl Acad Sci U S A**	107	14345-14350	2010
Aoyagi S, Hata H, Izumi K, Iitani MM, **Shimizu H**	Diagnostic pitfalls of using dermoscopic features to differentiate between malignant melanoma and pigmented seborrhoeic keratosis	**Acta Derm Venereol**	90	440-441	2010
Aoyagi S, Hata H, Homma E, **Shimizu H**	Controlling the histological margin for non-melanoma skin cancer conveniently using a double-bladed scalpel.	**J Surg Oncol**	101	175-179	2010
Akiyama M, Sakai K, Yanagi T, Tabata N, Yamada M, **Shimizu H**	Partially disturbed lamellar granule secretion in mild congenital ichthyosiform erythroderma with ALOX12B mutations.	**Br J Dermatol**	163	201-204	2010
Akiyama M, Sakai K, Yanagi T, Fukushima S, Ihn H, Hitomi K, **Shimizu H**	Transglutaminase1 preferred substrate peptide K5 is an efficient tool in diagnosis of lamellar ichthyosis.	**Am J Pathol**	176	1592-1599	2010
Abe R, Yamagishi S, Fujita Y, Hoshina D, Sasaki M, Nakamura K, Matsui T, Shimizu T, Bucala R, **Shimizu H**	Topical application of anti-angiogenic peptides based on pigment epithelium-derived factor can improve psoriasis.	**J Dermatol Sci**	57	183-191	2010
Akiyama M	ABCA12 mutations and autosomal recessive congenital ichthyosis: A review of genotype/phenotype correlations and of pathogenetic concepts. (Review article)	**Hum Mutation**	31	1090-1096	2010
Mori T, Ishida K, Mukumoto S, Yamada Y, Imokawa G, Kabashima K, Kobayashi M, Bito T, Nakamura M, Ogasawara K, **Tokura Y**	Comparison of skin barrier function and sensory nerve electric currentperception threshold between IgE-high extrinsic and IgE-normal intrinsic types of atopic dermatitis.	**Br J Dermatol**	162	83-90	2010

Yanagi T, Shimizu T, Kodama K, Nemoto-Hasebe I, Kasai M, **Shimizu H**	CD30-positive primary cutaneous anaplastic large-cell lymphoma and definite squamous cell carcinoma.	**Clin Exp Dermatol**	34	e293-294	2009
Ujiie H, **Akiyama M**, Osawa R, Shida S, Aoyagi S, **Shimizu H**	Bloody nipple discharge in an infant	**Arch Dermatol**	145	1068-1069	2009
Uematsu R, Shinohara Y, Nakagawa H, Kurogochi M, Furukawa J, Miura Y, **Akiyama M**, **Shimizu H**, Nishimura S	Glycosylation specific for adhesion molecules in epidermis and its receptor revealed by glycoform-focused reverse genomics.	**Mol Cell Proteomics**	8	232-244,	2009
Suzuki K, Yamaguchi Y, Villacorte M, Mihara K, **Akiyama M**, **Shimizu H**, Taketo MM, Nakagata N, Tsukiyama T, Yamaguchi TP, Birchmeier W, Kato S, Yamada G	Embryonic hair follicle fate change by augmented {beta}-catenin through Shh and Bmp signaling.	**Development**	136	367-372	2009
Shinkuma S, **Akiyama M**, Torii-Saito N, Natsuga K, Tateishi Y, Ito K, Hirota J, Shimizu Y, Shichinohe T, **Shimizu H**	Pemphigus foliaceus associated with oesophageal cancer.	**J Eur Acad Dermatol Venereol**	23	473-474	2009
Shinkuma S, Abe R, Nishimura M, Natsuga K, Fujita Y, **Nomura T**, Nishie W, **Shimizu H**	Secondary syphilis mimicking warts in an HIV-positive patient.	**Sex Transm Infect**	85	484	2009
Sakai K, **Akiyama M**, Yanagi T, McMillan JR, Suzuki T, Tsukamoto K, Sugiyama H, Hatano Y, Hayashitani M, Takamori K, Nakashima K, **Shimizu H**	ABCA12 is a major causative gene for non-bullous congenital ichthyosiform erythroderma.	**J Invest Dermatol**	129	2306-2309	2009
Qiao H, Shibaki A, Long HA, Wang G, Li Q, Nishie W, Abe R, **Akiyama M**, **Shimizu H**, McMillan JR	Collagen XVII participates in keratinocyte adhesion to collagen IV, and in p38MAPK-dependent migration and cell signaling.	**J Invest Dermatol**	129	2288-2295	2009
Osawa R, **Akiyama M**, Yamanaka Y, Ujiie H, Nemoto-Hasebe I, Takeda A, Yanagi T, **Shimizu H**	A novel PTPN11 missense mutation in a patient with LEOPARD syndrome.	**Br J Dermatol**	161	1202-1204	2009

Nomura Y, Nishie W, Shibaki A, Ibata M, **Shimizu H**	Disseminated cutaneous Mycobacterium kansasii infection in an patient infected with the human immunodeficiency virus.	**Clin Exp Dermatol**	34	625-626,	2009
Nomura Y, **Akiyama M**, Nishie W, **Shimizu H**	Progressive Refractory Ulcer of the Nipple: A Quiz.	**Acta Derm Venereol**	89	445-447	2009
Nomura Y, Abe M, Natsuga K, Moriuchi R, Kawasaki H, Mayuzumi M, Yasuoka A, **Shimizu H**	Widespread keratosis follicularis squamosa.	**Clin Exp Dermatol**	34	519-520	2009
Nishie W, Sawamura D, Natsuga K, Shinkuma S, Goto M, Shibaki A, Ujiie H, Olasz E, Yancey KB, **Shimizu H**	A novel humanized neonatal autoimmune blistering skin disease model induced by maternally transferred antibodies.	**J Immunol**	183	4088-4093	2009
Nemoto-Hasebe I, **Akiyama M**, Kudo S, Ishiko A, Tanaka A, **Arita K**, **Shimizu H**	Novel mutation p.Gly59Arg in GJB6 encoding connexin 30 underlies palmoplantar keratoderma with pseudoainhum, knuckle pads and hearing loss.	**Br J Dermatol**	161	452-455	2009
Kanda M, Natsuga K, Nishie W, **Akiyama M**, Nagasaki A, Shimizu T, **Shimizu H**	Morphological and genetic analysis of steatocystoma multiplex in an Asian family with pachyonychia congenita type 2 harbouring a KRT17 missense mutation.	**Br J Dermatol**	160	465-468	2009
Kawabori M, Kuroda S, Nakayama N, Kenmotsu Y, **Shimizu H**, Tanino M, Iwasaki Y	Spontaneous giant aneurysm of the superficial temporal artery: case report.	**Neurol Med Chir** (Tokyo)	49	198-201	2009
Ito K, Sawamura D, Goto M, Nakamura H, Nishie W, Sakai K, Natsuga K, Shinkuma S, Shibaki A, Uitto J, Denton CP, Nakajima O, **Akiyama M**, **Shimizu H**	Keratinocyte-/fibroblast-targeted rescue of Col7a1-disrupted mice and generation of an exact dystrophic epidermolysis bullosa model using a human COL7A1 mutation.	**Am J Pathol**	175	2508-2517	2009
Inomata K, Aoto T, Binh NT, Okamoto N, Tanimura S, Wakayama T, Iseki S, Hara E, Masunaga T, **Shimizu H**, Nishimura EK	Genotoxic stress abrogates renewal of melanocyte stem cells by triggering their differentiation.	**Cell**	137	1088-1099	2009

Inokuma D, Sawamura D, Shibaki A, Abe R, **Shimizu H**	Scleroedema adultorum associated with sarcoidosis	**Clin Exp Dermatol**	34	e428-429	2009
Inokuma D, Kodama K, Natsuga K, Kasai M, Abe M, Nishie W, Abe R, Hashimoto T, **Shimizu H**	Autoantibodies against type XVII collagen C-terminal domain in a patient with bullous pemphigoid associated with psoriasis vulgaris.	**Br J Dermatol**	160	451-454	2009
Inokuma D, Aoyagi S, Saito N, Iitani MM, Homma E, Hamasaka K, **Shimizu H**	Bowen's disease of the nail matrix presenting as melanonychia: detection of human papillomavirus type 56.	**Acta Derm Venereol**	89	638-639	2009
Imada K, Dainichi T, Yokomizo A, Tsunoda T, Song YH, Nagasaki A, Sawamura D, Nishie W, **Shimizu H**, Fukagawa S, Urabe K, Furue M, Hashimoto T, Naito S	Birt-Hogg-Dube syndrome with clear-cell and oncocytic renal tumour and trichoblastoma associated with a novel FLCN mutation.	**Br J Dermatol**	160	1350-1353	2009
Hsu CK, **Akiyama M**, Nemoto-Hasebe I, **Nomura T**, Sandilands A, Chao SC, Lee JY, Sheu HM, McLean WH, **Shimizu H**	Analysis of Taiwanese ichthyosis vulgaris families further demonstrates differences in FLG mutations between European and Asian populations.	**Br J Dermatol**	161	448-451	2009
Honda A, Abe R, Yoshihisa Y, Makino T, Matsunaga K, Nishihira J, **Shimizu H**, Shimizu T	Deficient deletion of apoptotic cells by macrophage migration inhibitory factor (MIF) overexpression accelerates photocarcinogenesis.	**Carcinogenesis**	30	1597-1605	2009
Hamasaka E, **Akiyama M**, Hata H, Aoyagi S, **Shimizu H**	Melanonychia caused by Stenotrophomonas maltophilia.	**Clin Exp Dermatol**	34	242-243	2009
Hamasaka A, Abe R, Koyama Y, Yoshioka N, Fujita Y, Hoshina D, Sasaki M, Hirasawa T, Onodera S, Ohshima S, Leng L, Bucala R, Nishihira J, Shimizu T, **Shimizu H**	DNA vaccination against macrophage migration inhibitory factor improves atopic dermatitis in murine models.	**J Allergy Clin Immunol**	124	90-99	2009

Goto-Ohguchi Y, Nishie W, **Akiyama M**, Tateishi Y, Aoyagi S, Tsuji-Abe Y, Sawamura D, Ishii N, Hashimoto T, **Shimizu H**	A severe and refractory case of anti-p200 pemphigoid resulting in multiple skin ulcers and scar formation.	**Dermatology**	218	265-271	2009
Bohgaki T, Atsumi T, Bohgaki M, Furusaki A, Kondo M, Sato-Matsumura KC, Abe R, Kataoka H, Horita T, Yasuda S, Amasaki Y, Nishio M, Sawada K, **Shimizu H**, Koike T	Immunological reconstitution after autologous hematopoietic stem cell transplantation in patients with systemic sclerosis: relationship between clinical benefits and intensity of immunosuppression.	**J Rheumatol**	36	1240-1248	2009
Asaka T, **Akiyama M**, Kitagawa Y, **Shimizu H**	Higher density of label-retaining cells in gingival epithelium.	**J Dermatol Sci**	55	132-134	2009
Asaka T, **Akiyama M**, Domon T, Nishie W, Natsuga K, Fujita Y, Abe R, Kitagawa Y, **Shimizu H**	Type XVII collagen is a key player in tooth enamel formation.	**Am J Pathol**	174	91-100	2009
Arita K, Abe R, Baba K, McGrath JA, **Akiyama M**, **Shimizu H**	A novel OSMR mutation in familial primary localized cutaneous amyloidosis in a Japanese family.	**J Dermatol Sci**	55	64-65	2009
Aoyagi S, Izumi K, Hata H, Kawasaki H, **Shimizu H**	Usefulness of real-time tissue elastography for detecting lymph-node metastases in squamous cell carcinoma.	**Clin Exp Dermatol**	34	e744-e747	2009
Aoyagi S, Hata H, Iitani MM, Homma E, Inokuma D, **Shimizu H**	Squamous cell carcinoma of the auricle with rhabdoid features.	**J Cutan Pathol**	36	919-921	2009
Ando S, Sato-Matsumura KC, Kasai M, Nemoto-Hasebe I, Hoshina D, Ohyama B, Hashimoto T, **Shimizu H**	Desmoglein1 and BP 180 ELISA indexes correlating with disease activity in a patient with coexisting pemphigus foliaceus and bullous pemphigoid.	**Clin Exp Dermatol**	34	e995-996	2009
Ando S, Abe R, Sasaki M, Murata J, Inokuma D, **Shimizu H**	Bone marrow-derived cells are not the origin of the cancer stem cells in ultraviolet-induced skin cancer.	**Am J Pathol**	174	595-601	2009

Amagai M, Ikeda S, **Shimizu H**, Iizuka H, Hanada K, Aiba S, Kaneko F, Izaki S, Tamaki K, Ikezawa Z, Takigawa M, Seishima M, Tanaka T, Miyachi Y, Katayama I, Horiguchi Y, Miyagawa S, Furukawa F, Iwatsuki K, Hide M, Tokura Y, Furue M, Hashimoto T, Ihn H, Fujiwara S, Nishikawa T, Ogawa H, Kitajima Y, Hashimoto K	A randomized double-blind trial of intravenous immunoglobulin for pemphigus.	**J Am Acad Dermatol**	60	595-603	2009
Akiyama M, Sakai K, Hayasaka K, Tabata N, Yamada M, Ujiie H, Shibaki A, **Shimizu H**	Conradi-Hunermann-Happle syndrome with abnormal lamellar granule contents.	**Br J Dermatol**	160	1335-1337	2009
Abe R, Yoshioka N, Murata J, Fujita Y, **Shimizu H**	Granulysin as a marker for early diagnosis of the Stevens-Johnson syndrome.	**Ann Intern Med**	151	514-515	2009

Ⅳ. 研究成果の刊行物・別刷

Japanese-Specific Filaggrin Gene Mutations in Japanese Patients Suffering from Atopic Eczema and Asthma

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TO THE EDITOR

Mutations in *FLG*, the gene encoding profilaggrin/filaggrin, are the underlying cause of ichthyosis vulgaris (OMIM 146700) and an important predisposing factor for atopic eczema (AE) (Sandilands *et al.*, 2007). *FLG* mutations are also significantly associated with asthma with AE mainly in the European population (Rodríguez *et al.*, 2009; van den Oord and Sheikh, 2010). The presence of population-specific *FLG* mutations has been reported in both the European and Asian races (Nomura *et al.*, 2007; Sandilands *et al.*, 2007). To clarify whether *FLG* mutations are a predisposing factor for asthma in the non-European population, we initially studied 172 Japanese AE patients (mean age, 24.8 ± 9.1 years) and 134 unrelated Japanese control individuals (healthy volunteers; mean age, 27.9 ± 6.0 years). All AE patients had been diagnosed based on widely recognized diagnostic criteria (Hanifin and Rajka, 1980). The majority of AE patients and control individuals were identical to those in a previous study (Nemoto-Hasebe *et al.*, 2010). In this AE cohort, 73 AE patients (mean age, 25.4 ± 8.9 years) experienced complications with asthma. Furthermore, we studied another Japanese asthma cohort (137 patients; mean age, 58.2 ± 16.9 years). Patients were considered asthmatic based on the presence of recurrent episodes of ≥2 of the three symptoms (coughing, wheezing, or dyspnea) associated with demonstrable reversible airflow limitation, either spontaneously or with an inhaled short-acting β2-agonist and/or increased airway responsiveness to methacholine (Isada *et al.*, 2010). Fully informed consent was obtained from the participants or their legal guardians for this study. This study had been approved by the Ethical Committee at Hokkaido University Graduate School of Medicine and was conducted according to the Declaration of Helsinki Principles.

FLG mutation screening revealed that 27.4% of patients in our Japanese AE complicated with asthma case series carried one or more of the eight *FLG* mutations (combined minor allele frequency of 0.151, $n=146$) (Table 1). Conversely, 26.3% of Japanese AE patients without asthma carried one or more of the eight *FLG* mutations (combined minor allele frequency of 0.147, $n=198$). The *FLG* variants are also carried by 3.7% of Japanese control individuals (combined minor allele frequency of 0.019, $n=268$). We found that all compound heterozygous mutations were present in *trans* by observing transmission or haplotype analysis (Nomura *et al.*, 2007, 2008). There is a statistically significant association between the eight *FLG* mutations and AE with asthma, and between the eight *FLG* mutations and AE without asthma (Table 1). Moreover, AE complicated with asthma manifested in heterozygous carriers of *FLG* mutations with an odds ratio for AE and asthma of 9.74 (95% confidence interval 3.47–27.32), suggesting a relationship between *FLG* mutations and AE with asthma.

In the Japanese general asthma cohort, 8.0% of the asthma patients carried one or more of the eight *FLG* mutations (combined minor allele frequency of 0.04, $n=274$) (Table 2). Whereas, of the Japanese patients with asthma complicated by AE, 22.2% carried one or more of the *FLG* mutations (combined minor allele frequency of 0.11, $n=36$). In contrast, 5.9% of asthma patients without AE carried one or more of the *FLG* mutations (combined minor allele frequency of 0.03, $n=238$). There was a statistically significant association between the eight *FLG* mutations and asthma with AE (Table 2). There was no statistically significant association between the *FLG* mutations and entire asthma patients, nor between *FLG* mutations and asthma without AE. We cannot exclude the possibility that this lack of significant association is due to the small number of the patients included in this study. We used the same control set for both case-controlled studies. Thus, strictly speaking, there is no independent replication for the control group.

Recent meta-analysis revealed that *FLG* mutations are significantly associated with asthma in the European population and there are especially, strong effects observed for *FLG* mutations for the compound phenotype, asthma in addition to eczema (Rodríguez *et al.*, 2009; van den Oord and Sheikh, 2010). In contrast, there appeared to be no association of *FLG* mutations with asthma in the absence of eczema (Rodríguez *et al.*, 2009; van den Oord and Sheikh, 2010).

This Japanese cohort has a completely different *FLG* mutation spectrum from those in the European and the North American populations. However, our results clearly confirm the strong association of *FLG* mutations with our Japanese cohort of AE patients with asthma complications, and the association of *FLG* mutations and asthma patients with AE complications, for the first time outside Europe or North America. Conversely, this study showed no significant correlation between general asthma patients and *FLG* mutations, suggesting that atopic asthma patients associated with *FLG* mutations are a minority among general asthma patients. The frequency of heterozygous, compound heterozygous, and homozygous *FLG* mutation carriers

Abbreviation: AE, atopic eczema

Table 1. Atopic eczema case–control association analysis for *FLG* null variants in Japan

	R501X		3321delA		S1695X		Q1701X		S2554X		S2889X		S3296X		K4022X		Combined			
Genotype	Con	AE	Con	AE	Con	AE	Con	AE	Con	AE	Con	AE	Con	AE	Con	AE	Con	AE (total)	AE (asthma+)	AE (asthma−)
AA	134	172	133	163	133	172	134	169	133	162	132	152	134	166	134	169	129	126	53	73
Aa	0	0	1	9	1	0	0	3	1	10	2	20	0	6	0	3	5	41	18	23
aa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	5[1]	2	3
Total	134	172	134	172	134	172	134	172	134	172	134	172	134	172	134	172	134	172	73	99

Abbreviations: AE, atopic eczema; CI, confidence interval; Con, healthy control; OR, odds ratio.
For combined genotype: AE+asthma, exact *P*-value of Pearson χ^2-test=1.909×10^{-6}, OR and 95% CI for dominant models (AA vs aX)=9.737 (3.473–27.322); AE−asthma, exact *P*-value of Pearson χ^2-test=7.189×10^{-7}, OR and 95% CI for dominant models (AA vs aX)=9.191 (3.383–24.938); all AE, exact *P*-value of Pearson χ^2-test=1.189×10^{-7}, OR and 95% CI for dominant models (AA vs aX)=9.416 (3.625–24.450).
[1]All the five patients were compound heterozygotes for minor alleles.

Table 2. Asthma case–control association analysis for *FLG* null variants in Japan

	R501X		3321delA		S1695X		Q1701X		S2554X		S2889X		S3296X		K4022X		Combined			
Genotype	Con	Asthma	Con	Asthma	Con	Asthma	Con	Asthma	Con	Asthma	Con	Asthma	Con	Asthma	Con	Asthma	Con	Asthma (total)	Asthma (AE+)	Asthma (AE−)
AA	134	137	133	137	133	137	134	137	133	133	132	132	134	136	134	136	129	126	14	112
Aa	0	0	1	0	1	0	0	0	1	4	2	5	0	1	0	1	5	11	4	7
aa	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Total	134	137	134	137	134	137	134	137	134	137	134	137	134	137	134	137	134	137	18	119

Abbreviations: AE, atopic eczema; CI, confidence interval; Con, healthy control; OR, odds ratio.
For combined genotype: asthma+AE, exact *P*-value of Pearson χ^2-test=0.0122, OR and 95% CI for dominant models (AA vs aX)=7.3692 (1.7715–30.6748); asthma−AE, exact *P*-value of Pearson χ^2-test=0.5563, OR and 95% CI for dominant models (AA vs aX)=1.6124 (0.4979–5.2219); all asthma, exact *P*-value of Pearson χ^2-test=0.1968, OR and 95% CI for dominant models (AA vs aX)=2.2523 (0.7609–6.6667).

observed in our Japanese controls was only 3.7%, which was much lower than that seen in European general population, where it is approximately 7.5%. This suggested that there may be further mutations yet to be discovered in the Japanese. As we have sequenced more than 40 Japanese families with ichthyosis vulgaris, there is now little possibility that further highly prevalent mutations will be found in the Japanese population. However, it is still possible that there might be multiple, further low-frequency *FLG* mutations discovered in the Japanese population. In addition, because of the relatively small sample size of this genetic study, further replication in association studies will be required for *FLG* mutations and asthma in Japan.

In our cohorts, serum IgE levels were extremely high (median, 3141.9 IU ml^{-1}; 25th–75th percentiles, 1276.0–9753.0 IU ml^{-1}) in AE patients with asthma ($n=73$) in the AE cohort, compared with that in total asthma patients (median, 156.0 IU ml^{-1}; 25th–75th percentiles, 71.05–441.45 IU ml^{-1}, $n=137$) in the asthma cohort. These findings suggest that extrinsic allergic sensitization might have an important role in atopic asthma pathogenesis. Recent studies hypothesized skin barrier defects caused by *FLG* mutation(s) allow allergens to penetrate the skin, resulting in initiation of further immune response and leading to the development of systemic allergies, including atopic asthma (Fallon *et al.*, 2009). In patients with asthma that also harbor *FLG* mutations, we could not exclude the possibility that the systemic effects of early eczema might simply influence airway responsiveness (Henderson *et al.*, 2008).

CONFLICT OF INTEREST

Irwin McLean has filed patents relating to genetic testing and therapy development aimed at the filaggrin gene.

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See related commentary on pg 2703

RNase 7 Protects Healthy Skin from *Staphylococcus aureus* Colonization

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TO THE EDITOR

The Gram-positive bacterium *Staphylococcus aureus* is an important pathogen that causes various skin infections (Miller and Kaplan, 2009). However, healthy skin is usually not infected by *S. aureus,* despite the high carrier rates in the normal population (Noble, 1998). This suggests that the cutaneous defense system has the capacity to effectively control the growth of *S. aureus*. There is increasing evidence that antimicrobial proteins are important effectors of the cutaneous defense system (Harder *et al.*, 2007). A recent study reported that keratinocytes contribute to cutaneous innate defense against *S. aureus* through the production of human β-defensin-3 (Kisich *et al.*, 2007). In addition to human β-defensin-3,, other antimicrobial proteins may also participate in cutaneous defense against *S. aureus*. One candidate is RNase 7, a potent antimicrobial ribonuclease that is highly expressed in healthy skin (Harder and Schröder, 2002; Köten *et al.*, 2009).

To investigate the hypothesis that RNase 7 may contribute to protect healthy skin from *S. aureus* colonization, we first incubated natural RNase 7 isolated from stratum corneum skin extracts (Harder and Schröder, 2002) with *S. aureus* (ATCC 6538). In concordance with our initial report about RNase 7 (Harder and Schröder, 2002), we verified that RNase 7 exhibited a high killing activity against *S. aureus* (lethal dose of 90% $= 3–6\ \mu g\ ml^{-1}$).

Recently, we reported a moderate induction of RNase 7 mRNA expression in primary keratinocytes treated with heat-killed *S. aureus* (Harder and Schröder, 2002). To assess the induction of RNase 7 by *S. aureus* in the

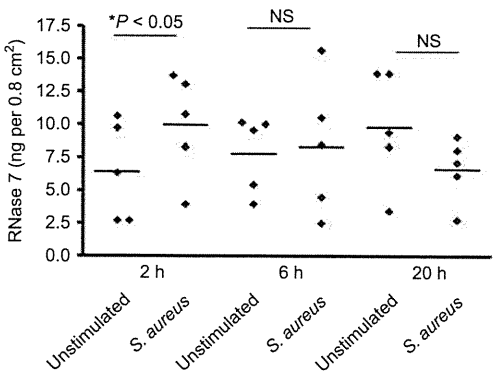

Figure 1. Induced secretion of RNase 7 on the skin surface on treatment with living *S. aureus*. Defined areas (0.8 cm^2) of skin explants derived from plastic surgery were incubated with or without approximately 1,000 colony-forming units of *S. aureus* (ATCC 6538) in 100 μl of sodium phosphate buffer. After 2, 6, and 20 hours, the concentration of secreted RNase 7 was determined by ELISA. Stimulation with *S. aureus* for 2 hours revealed a significant induction as compared with the unstimulated control after 2 hours (*$P<0.05$, Student's *t*-test; n.s. = not significant). Data shown are means of triplicates of five skin explants derived from five donors.

coexistent proliferative epidermal lesions, such as condyloma acuminatum, Bowen's disease, and squamous cell carcinoma, but not in adjacent EMPD areas [3,9,10]. Therefore, HPV infection in these cases is more likely coincidental than causal in the pathogenesis of EMPD, although the precise relationship still needs to be elucidated.

Our findings provide further evidence that HPV infection is unlikely to contribute to the carcinogenesis of EMPD. However, further investigation is required to determine whether or not there is an association between EMPD and other types of HPV that were not detected by the methods used in this study.

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Letter to the Editor

Chromosome 11q13.5 variant: No association with atopic eczema in the Japanese population

Dear Sir,

A single nucleotide polymorphism (SNP) on chromosome 11q13.5 [rs7927894] has been attracting great attention since Esparza-Gordillo et al. [1] reported highly significant association of a common variant of rs7927894 with atopic eczema (AE) in the German population. In the report, approximately 13% of individuals are homozygous for the SNP and their risk of developing AE is 1.47 times that of noncarriers. Very recently, O'Regan et al. [2] further published interesting results on the association between rs7927894 and AE in a collection of Irish children with moderate-to-severe AE and Irish controls. The association between rs7927894 and AE was replicated in the Irish population ($p = 0.0025$, Chi-square test; odds ratio (OR) = 1.27, 95% confidence interval (CI) 1.09–1.49). Additional analyses performed to test the statistical significance of the rs7927894 SNP having controlled for the presence/absence of the strongly significant *FLG* null genotype indicated that rs7927894 still shows a statistically significant effect ($p = 0.0025$) with an OR of 1.22 (95% CI 1.02–1.26) [2]. Tests for interaction between each of the *FLG* and rs7927894 risk alleles showed no evidence of statistically significant epistatic effects [2]. The rs7927894 association was independent of the well-established *FLG* risk alleles and may be multiplicative in its effects.

In order to clarify whether this common variant is associated with AE also in the Japanese population or not, we evaluated the association between rs7927894 and AE in an cohort of 194 Japanese AE patients we had collected to date and 113 unrelated Japanese control individuals. All the AE patients had been diagnosed with AE based on widely recognized diagnostic criteria [3] or their parents reported a dermatologist's diagnosis of AE (at least once). Majority of AE patients and control individuals were identical to those in a previous study [4]. Using genomic DNA, AE patients and control individuals were screened for the variant allele of rs7927894 on chromosome 11q13, by direct DNA sequencing. In addition, the AE patients and the control individuals were screened for eight *FLG* mutations previously identified in the Japanese population, by restriction enzyme digestion, fluorescent PCR and/or direct DNA sequencing as described previously [4,5].

Case–control association analyses were performed for the variant using Fisher's exact test. In addition, we performed case–control statistical analysis for the common variant allele of rs7927894 after stratification for *FLG* mutations. The rs7927894 on chromosome 11q13 genotype data in the Japanese AE case series and ethnically matched population control series are summarized in Table 1. All alleles were observed to be in normal Hardy–Weinberg equilibrium.

Here we demonstrate that 22.7% and 1.5% of the patients in our Japanese AE case series are heterozygous and homozygous for rs7927894[T], respectively (combined rs7927894[T] allele frequency = 0.129, $n = 388$) (Table 1). rs7927894[T] is also carried by 23.0% of the Japanese control individuals (combined minor allele frequency = 0.115, $n = 226$). There is no statistically significant association between the rs7927894[T] and AE.

After stratification for *FLG* mutations previously identified in the Japanese population, 26.0% and 4.0% of our Japanese AE case series with *FLG* mutations are heterozygous and homozygous for rs7927894[T] (combined rs7927894[T] allele frequency = 0.17, $n = 100$). 21.5% and 0.7% of the Japanese AE patients without *FLG* mutations are heterozygous and homozygous for rs7927894[T] (combined rs7927894[T] allele frequency = 0.11, $n = 288$). There is no statistically significant association between the rs7927894[T] and AE without *FLG* mutations or rs7927894[T] and AE with *FLG* mutations (Fisher's exact test $p = 0.338$). Furthermore, interaction

Table 1
Results of the 11q13.5 SNP and the prevalent *FLG* mutations in 194 Japanese eczema cases and 113 individuals from Japanese control population.

	Eczema cases			Control		
	Total	*FLG* (+)	*FLG* (−)	Total	*FLG* (+)	*FLG* (−)
C/C	147 (75.8%)	35 (70.0%)	112 (77.8%)	87 (77.0%)	2 (50.0%)	85 (78.0%)
C/T	44 (22.7%)	13 (26.0%)	31 (21.5%)	26 (23.0%)	2 (50.0%)	24 (22.0%)
T/T	3 (1.5%)	2 (4.0%)	1 (0.7%)	0 (0%)	0 (0%)	0 (0%)
Total	194	50	144	113	4	109

FLG (+), with *FLG* mutation(s); *FLG* (−), without any *FLG* mutation. Combined rs7927894[T] allele frequency, 0.129 (AE patients, $n = 388$); 0.115 (control individuals, $n = 226$); 0.17 (AE patients with *FLG* mutation(s), $n = 100$); 0.115 (AE patients without *FLG* mutation(s), $n = 288$).

Table 2
Cross-classification of genotypes of rs7927894 and *FLG* used for the interaction analysis.

Genotype		Cases			Controls		
		rs7927894			rs7927894		
		AA	AA	Aa	AA	Aa	aa
R501X	AA	147	44	3	87	26	0
	Aa	0	0	0	0	0	0
	aa	0	0	0	0	0	0
3321delA	AA	141	42	2	87	25	0
	Aa	6	2	1	0	1	0
	aa	0	0	0	0	0	0
S1695X	AA	147	44	3	86	26	0
	Aa	0	0	0	1	0	0
	aa	0	0	0	0	0	0
Q1701X	AA	144	44	3	87	26	0
	Aa	3	0	0	0	0	0
	aa	0	0	0	0	0	0
S2554X	AA	141	41	3	87	26	0
	Aa	6	3	0	0	0	0
	aa	0	0	0	0	0	0
S2889X	AA	133	36	2	86	25	0
	Aa	14	8	1	1	1	0
	Aa	0	0	0	0	0	0
S3296X	AA	141	43	3	87	26	0
	Aa	6	1	0	0	0	0
	Aa	0	0	0	0	0	0
K4022X	AA	144	43	3	87	26	0
	Aa	3	1	0	0	0	0
	Aa	0	0	0	0	0	0
Combined *FLG* null genotype	AA	109	31	1	85	24	0
	Aa	35	13	2	2	2	0
	Aa	0	0	0	0	0	0

AA, wild-type homozygous individuals for each genetic variant; Aa, wild-type/mutant heterozygous individuals; aa, individuals who are homozygous for each of the genetic variants tested.

between each of the *FLG* and rs7927894 risk alleles based on the cross-classification of genotypes in Table 2 showed no apparent epistatic effects.

rs7927894 is located in an intergenic region 38 kb downstream of *C11orf30* (chromosome 11 open reading frame 30) and 68 kb upstream of *LRRC322* (leucine rich repeat containing 32). Both *C11orf30* and *LRRC322* are ubiquitously expressed including skin and peripheral blood lymphocytes [1]. By genome-wide association study for global mRNA expression in lymphoblastoid cell lines from asthmatic children, there was no evidence for a *cis*-regulatory effect of rs7927894 [6]. Thus, regulatory role of rs7927894 on *C11orf30* and *LRRC322* gene expression is questionable. However, we cannot exclude the possibility of a pathogenetic link of rs7927894 to atopic eczema via *C11orf30* and *LRRC322* gene expression in the skin.

Our case–control study in the Japanese population did not confirm the result of Esparza-Gordillo et al. [1] or O'Regan et al. [2] that rs7927894 is at increased risk for AE. The association of rs7927894 with AE was reported in the European population, i.e. in the German population by Esparza-Gordillo et al. [1] and in the Irish population by O'Regan et al. [2]. The present data suggest that the association of rs7927894 with AE established in the European populations is not in the Asian populations, at least in the Japanese population.

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EXPERT REVIEWS

Update on filaggrin mutations and atopic dermatitis

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Skin serves as a protective barrier against invasion by pathogens and harmful antigenic particles. Filaggrin is a key structural protein that facilitates terminal differentiation of the keratinocytes and formation of the skin barrier. Since the establishment of a sequencing method for the entire filaggrin gene (*FLG*) in 2006, approximately 40 loss-of-function *FLG* mutations have been identified in patients with ichthyosis vulgaris and/or atopic dermatitis (AD). Notably, there is a clear difference in filaggrin genetics between the European and Asian races. Overall, approximately 25–50% of AD patients have been found to harbor filaggrin mutations as a predisposing factor. In addition, filaggrin mutations are significantly associated with asthma. The restoration of skin barrier function seems a feasible and promising strategy for prophylactic treatment of AD patients with *FLG* mutations. This article reviews the discovery of filaggrin mutations; their association with AD, asthma and other atopic diseases; and *FLG*-related potential treatment strategies.

KEYWORDS: atopic dermatitis • eczema • filaggrin • *FLG* • ichthyosis vulgaris

Filaggrin, which is processed from profilaggrin, is a key structural protein that facilitates terminal differentiation of the epidermis and formation of the skin barrier. Mutations in *FLG*, the gene encoding filaggrin, have been identified as the cause of ichthyosis vulgaris (IV), a relatively common genetic keratinization disorder that is clinically characterized by scaling, especially on the extensor limbs, and palmoplantar hyperlinearity [1–3]. In 2006, the molecular basis and full sequencing of *FLG* were established [4]. Approximately 40 *FLG* mutations have been reported, and the prevalent ones are distinct in different populations [5]. Recent studies have shown that *FLG* mutations are also a key predisposing factor for atopic dermatitis (AD) [6], and for other atopic disorders, including asthma and allergic rhinitis [7]. This article reviews the discovery of filaggrin mutations and its association with AD.

Skin barrier function

The primary function of the skin is to act as a protective barrier against invasion by harmful organisms, such as bacteria, viruses, fungi and other antigenic particles. Keratinocytes are the principal cells within the epidermis. The terminal differentiation of keratinocytes (FIGURE 1) results in the formation of an impenetrable barrier (the horny layer) that is the uppermost layer of the epidermis. The successive stages of keratinocytic differentiation in the epidermal layers are in the basal cell, spinous cell and granular cell layers (FIGURE 2). While spinous cells differentiate into granular cells, they begin to accumulate keratinocyte-specific proteins involved in terminal differentiation of the horny layer. There are three major components in the skin barrier of the horny layer: intercellular lipid layers; the cornified cell envelope; and the keratin network and keratohyaline granules [8]. Genetic defects in any of these components may result in various dermatoses, such as ichthyoses, which are usually characterized by dry, thickened, scaly or flaky skin ('ichthyosis' comes from the Ancient Greek word *'ichthys'*, meaning 'fish').

The keratin filament network is an important basic structure for maintaining the integrity and dimensions of the cornified cell, and the degraded product of keratohyalin granules – that is, filaggrin – aggregates keratin filaments in apoptosed keratinocytes into bundles and promotes the flattening of dead-cell remnants [9–12]. This layer of collapsed cells, which is reinforced by other structural proteins, forms an effective barrier against external allergens in normal skin.

Filaggrin

The term 'filaggrin' (a shortening of the phrase 'filament aggregation protein') first appeared in 1981 to describe a class of structural proteins that

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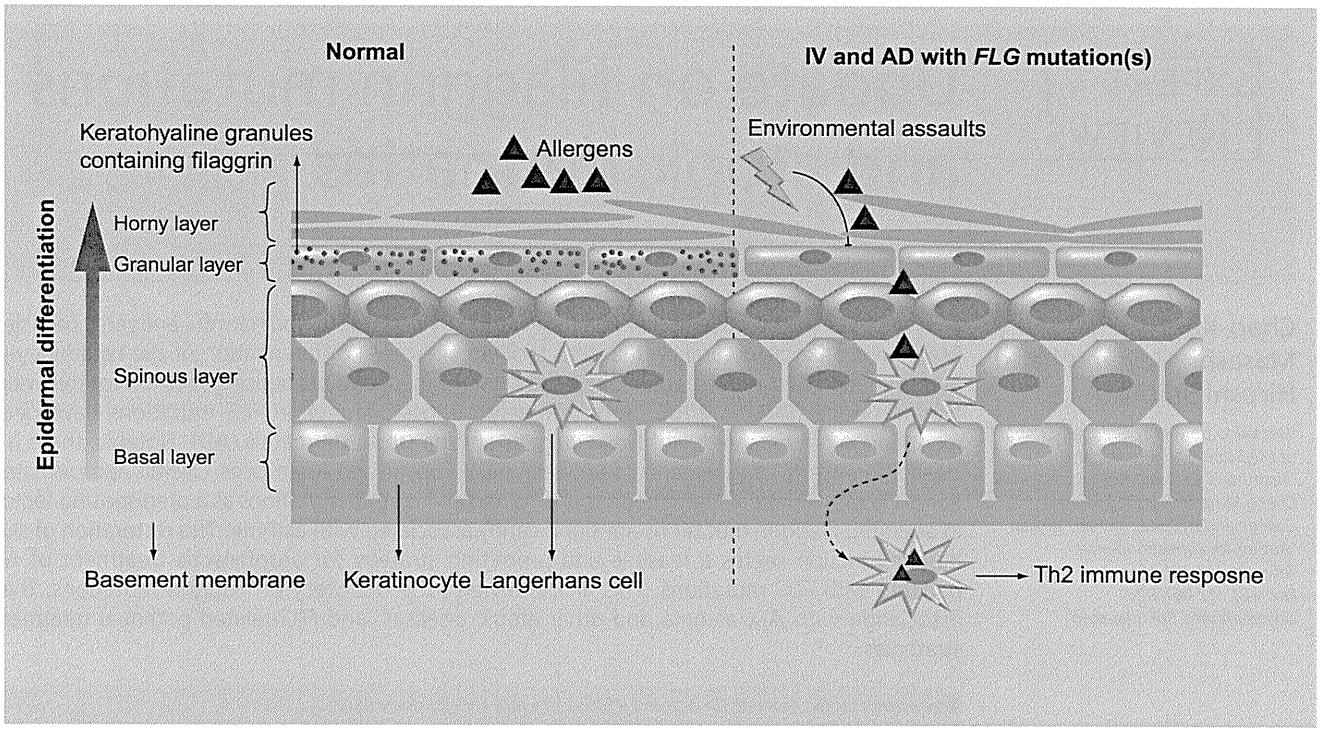

Figure 1. Anatomy and function of human skin barrier. The major cell population in the epidermis is keratinocytes, which undergo progressive differentiation from the basal layer to the granular layer, spinous layer and horny layer. In the granular layer, keratohyaline granules composed of profilaggrin predominate. Upon terminal differentiation of keratinocytes, the degraded product, filaggrin, aggregates keratin filaments and flattens the keratinocytes to form an effective barrier against external allergens in normal skin. In ichthyosis vulgaris and atopic dermatitis with *FLG* mutation, there is a reduction or complete absence of filaggrin. The defective skin barrier allows the external antigens to penetrate into the epidermis and interact with antigen-presenting cells, Langerhans cells and dermal dendritic cells, which might further initiate Th2 immune response and lead to atopic disorders.
Modified from [2].

are isolated from the horny layer [9]. Filaggrin is initially synthesized as profilaggrin, an approximately 500-kDa, highly phosphorylated, histidine-rich polypeptide that consists of an amino-terminal S100 calcium-binding domain, a B-domain and two imperfect filaggrin-repeat domains flanking 10–12 essentially identical filaggrin repeats, as well as of a carboxy-terminal domain (Figure 2) [13,14]. During the post-translational processing of profilaggrin, the 10–12 individual 37-kDa filaggrin polypeptides cleave proteolytically and then dephosphorylate. As mentioned above, the liberated filaggrin subsequently and highly efficiently aggregates the keratin filaments, which causes the keratinocytes in the stratum corneum to collapse [9,12]. Filaggrin subsequently degrades into amino acids, which act in retaining epidermal moisture [12,15]. The aspartate-specific protease caspase 14 plays an important role in the cleavage of profilaggrin [16]. Caspase-14-knockout mice show an abnormal accumulation of filaggrin fragments with a low molecular mass (12–15 kDa) within the stratum corneum [17]. Filaggrin is a key protein during terminal differentiation, and it is essential for the formation of an intact, protective and properly moisturized skin barrier [8,12].

Filaggrin loss-of-function mutations in ichthyosis vulgaris

Ichthyosis vulgaris (OMIM 146700) is a common inherited skin disorder that is estimated to affect one in 250 individuals. IV is characterized by generalized dry and scaly skin prominent on the extensor surfaces of limbs, and is associated with palmoplantar hyperlinearity (Figure 3) [1,4]. Histologically, IV is characterized by a decrease in the size and number of keratohyaline granules in the granular layer, or in their complete absence there (Figure 3) [1,18]. An association between IV and profilaggrin had long been suspected, but the gene that encodes profilaggrin, *FLG*, proved to be technically challenging to sequence. *FLG* resides on human chromosome 1q21 within the so-called epidermal-differentiation complex (EDC). The EDC contains an area of 1.62 megabases harboring more than 70 genes that are expressed during terminal differentiation of keratinocytes [19,20]. These EDC proteins, such as loricrin, involucrin, small protein-rich proteins and late envelop proteins, share similar important sequences, and phylogenic study suggests that these proteins derive from a common ancestor [21]. Of these EDC proteins, filaggrin is the key member.

The initiation codon of the *FLG* gene is in exon 2, and most of the profilaggrin protein is encoded by exon 3 (Figure 2). Exon 3 is extremely large (>12 kb) and it encodes most of the profilaggrin polypeptides, with almost completely homologous 10, 11 or 12 repeats. There exist polymorphic variations in the number of filaggrin repeats. Some individuals have a duplication of the