



Invited review article

New concept of the pathogenesis of atopic dermatitis: Interplay among the barrier, allergy, and pruritus as a trinity

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ABSTRACT

Atopic dermatitis (AD) is a common skin condition, characterized by a complex, heterogeneous pathogenesis, including skin barrier dysfunctions, allergy/immunology, and pruritus. When the skin barrier is disrupted by, for example, the filaggrin gene mutation and/or environmental factors, the skin is predisposed to being penetrated by external stimuli. Foreign antigens can be subdivided into two subsets by size: haptens (including metals) and protein antigens. It is known that a single hapten challenge provokes Th1 initially, but that repeated elicitation with haptens results in a shift toward Th2-dominated responses. On the other hand, exposure to protein antigens directly induces Th2-dominant conditions via the thymic stromal lymphopoietin (TSLP) receptor on Langerhans cells. Recently, it has been revealed that Th2 cells produce IL-31, which provokes pruritus, and that Th2 cytokines decrease filaggrin expressions by keratinocytes. These findings suggest that Th2 conditions lead to pruritus and barrier dysfunctions. In this review, we will examine the highly complex interplay among skin barrier abnormality, allergy/immunology, and pruritus as a trinity in the development of AD.

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1. Introduction

Atopic dermatitis (AD) is a relapsing chronic inflammatory skin disease characterized by eczematous skin lesions and intense pruritus [1]. AD is one of the most frequent chronic inflammatory skin diseases and its prevalence is increasing, affecting at least 15% of children and 2–10% of adults in industrialized countries [2]. Patients with AD often have other allergic diseases, including food allergies, asthma, and allergic rhinitis [2]; these often begin early in life and progress in a typical fashion—this is called the allergic (or atopic) march [3]. The skin is an active immune system organ that influences systemic immunity [4].

AD can be categorized into two types: extrinsic or intrinsic. Extrinsic or allergic AD exhibits high total serum IgE levels and the presence of specific IgE for environmental and food allergens, whereas intrinsic or non-allergic AD exhibits normal total IgE values and the absence of specific IgE without filaggrin gene mutations [5]. The skin barrier is perturbed in the extrinsic, but not intrinsic type. The pathogenesis of AD has been attributed to a complex interaction of the environment and host susceptibility genes, altered skin barrier function, the immune system, and pruritus [6,7]. Thus far, each component has been studied independently; however, recent findings have suggested that they interact in a highly complex manner in the development of AD. In this review, we focus on the role of barrier functions, immune systems, and pruritus in AD, and we discuss their interplay in the development of AD.

2. Barrier functions in AD

2.1. Stratum corneum and tight junction

Outermost barriers are critical to avoid desiccation and to protect against foreign insults. Mammalian skin consists of two sets of barriers: stratum corneum and tight junctions (TJs). Thus far, at least four causes of xerosis have been considered: (1) decrease in skin ceramides [8], (ii) alterations of the stratum corneum pH [9], (iii) overexpression of the proteases, including kallikreins (KLKs) and chymases [10], and (iv) defect in *Filaggrin* (FLG) [11] (Fig. 1).

In Netherton syndrome, unregulated pH-sensitive KLK5 directly activates the proteinase-activated receptor (PAR)-2 and induces nuclear factor kappaB (NFκB)-mediated overexpression of thymic stromal lymphopoietin (TSLP), which aggravates AD-like skin lesions [10]. Consistently, a PAR-2 inhibitor abrogated the clinical

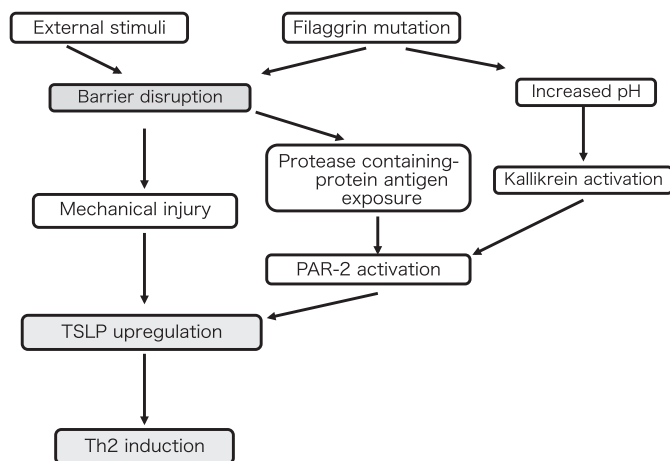


Fig. 1. Effect of barrier dysfunction on Th2 induction. Barrier dysfunction in line with filaggrin deficiency will lead to Th2 skewing conditions, which may play an important role in the development of AD.

manifestations in flaky tail (*Flg^{fl}*) mice, which is a mouse model of AD, as described below [12,13].

The role of TJs in AD remains unknown. A knockout study of claudin-1, a TJ-specific integral membrane protein, demonstrated that TJs function as paracellular diffusion barriers in mammalian epidermis [14]. In humans, lack of claudin-1 leads to ichthyosis with scalp hypotrichosis, scarring alopecia, neonatal sclerosing cholangitis, and leukocyte vacuolization (NISCH syndrome) [15], although a precise description of skin manifestation from the perspective of AD has not been reported.

One of the characteristics of AD is dry skin that affects both lesional and non-lesional skin areas [1]. Dry skin in AD parallels increased transepidermal water loss, which suggests that the skin barrier is disrupted in AD. It has long been thought that the barrier abnormality in AD is not merely an epiphenomenon, but is instead the initiator of its pathogenesis [16]. As a result of barrier disruption, the skin may permit the penetration of external stimuli, such as allergens (which may contain proteases), bacteria, and viruses [17].

2.2. Filaggrin

The direct evidence for a primary structural abnormality of the stratum corneum in AD is a recently discovered link between the incidence of AD and loss-of-function mutations in the gene encoding *FLG*. Individuals carrying the *FLG* null allele variants tend to develop AD [11]. *FLG* protein is localized in the granular layers of the epidermis (Fig. 2). Profilaggrin, a 400 kDa polyprotein, is the main component of keratohyalin granules [18,19]. In the differentiation of keratinocytes, profilaggrin is dephosphorylated and cleaved into 10–12 *FLG* molecules (with a molecular mass of 37 kDa in human and 27 kDa in mice), which aggregates in the keratin cytoskeleton system to form a dense protein-lipid matrix in humans [18]. These *FLG* monomers are further degraded into natural moisturizing factors (NMFs) by caspase 14, peptidylarginine deiminases, or bleomycin hydrolase. Filaggrin-derived NMFs are important to maintain skin hydration and low pH (Fig. 2). Intriguingly, it has recently been reported that intragenic copy number variation (20–24 copies in one person) within a *FLG* gene contributes to the risk of AD with a dose-dependent effect [20].

2.3. Filaggrin in mouse and human AD

Flaky tail (*Flg^{fl}*) mice, deficient in *Flg* and *matted* (*ma*), have been used to investigate the role of filaggrin in AD [21]. There have been four recent studies using *Flg^{fl}* mice as a model of filaggrin deficiency: Fallon et al. [22] used *Flg^{fl}* mice in which the *ma* mutation had been eliminated with four additional backcrosses to B6 mice, and the others used the commercially available *Flg^{fl}* mice [12,23,24].

The first report showed only a histological abnormality without clinical manifestations [22]. The second report demonstrated spontaneous eczematous skin lesions after 28 weeks of age [23]. The third report did not indicate any spontaneous dermatitis in *Flg^{fl}* mice [24]. The fourth report observed spontaneous dermatitis as early as five weeks of age with gradual exacerbation with age [12]. This time course differs from that of humans since most cases of AD resolve with age in humans. In addition, the cutaneous manifestations only occur in homozygous *Flg^{fl}* mice, but heterozygous carriers of a loss-of-function mutation of the *FLG* gene predispose humans to AD. The discrepancies among these results seem to be related to the presence or absence of the *ma* mutation and/or variation in the genetic backgrounds of the different strains used and to environmental factors. Recently, *Flg* null mutation mice have been generated that demonstrated that antigens penetrated the *Flg* null stratum corneum more efficiently, leading

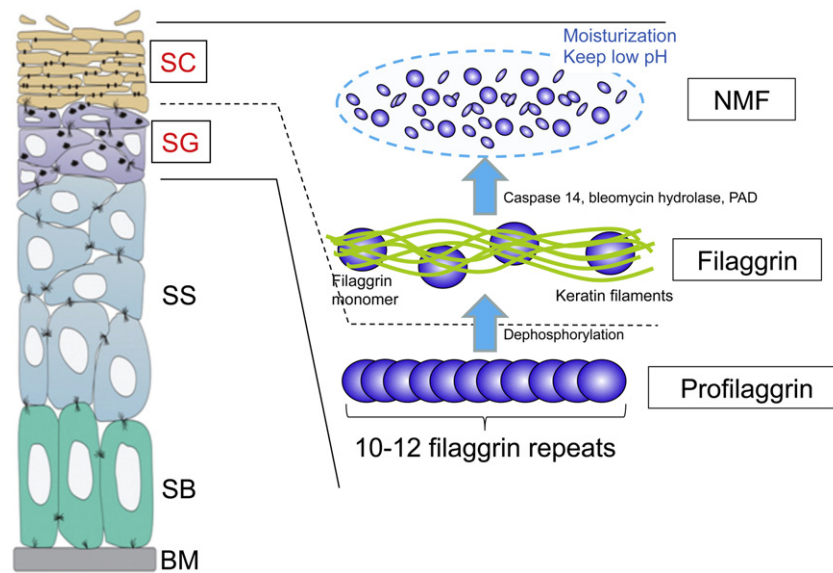


Fig. 2. Role of filaggrin. FLG protein is localized in the granular layers of the epidermis. Profilaggrin is the main component of keratohyalin granules. In the differentiation of keratinocytes, profilaggrin is dephosphorylated and cleaved into 10–12 FLG molecules, which aggregate in the keratin cytoskeleton system to form a dense protein-lipid matrix. These FLG monomers are further degraded into natural moisturizing factors, which are important to maintaining skin hydration and a low pH. SC, stratum corneum; SG, stratum granulosum; SS, stratum spinosum; SB, stratum basale; BM, basement membrane; NMF, natural moisturizing factor; PAD, peptidylarginine deiminase.

to enhanced responses in hapten-induced contact hypersensitivity (CHS) and higher serum levels of anti-ovalbumin IgG1 and IgE [25]. Intriguingly, these mice did not exhibit spontaneous AD-like skin lesions, which contrasts with the phenotype of *Flg^{fl}* mice.

3. Allergy/immunology

Human AD can be categorized into two types: extrinsic and intrinsic [5] (Table 1). Extrinsic or allergic AD exhibits a high total serum IgE levels and the presence of specific IgE for environmental and food allergens, whereas intrinsic or non-allergic AD exhibits normal total IgE values and the absence of specific IgE [5]. The skin barrier is perturbed in the extrinsic type, which increases the pre-existing pruritus, but not in the intrinsic type. *FLG* gene mutations are a feature of extrinsic, but not intrinsic, AD [26,27]. The intrinsic type is immunologically characterized by the lower expression of Th2 cytokines, interleukin (IL)-4, IL-5, and IL-13 in accordance with normal serum IgE levels, and the higher expression of the Th1 cytokine, interferon (IFN)- γ (Table 1). It has been suggested that intrinsic AD patients may not be sensitized to protein allergens, which induce Th2 responses, but that metals that may not induce IgE may be one of the causative allergen candidates [5,28].

3.1. Keratinocytes

Patients with AD (extrinsic AD) exhibit a compromised barrier function that leads to the activation of keratinocytes, which favors a Th2 bias. A wide array of cytokines and chemokines interact to

produce symptoms that are characteristic of AD. For example, barrier disruption promotes the production of thymus- and activation-regulated chemokine (TARC/CCL17) and macrophage-derived chemokine (MDC/CCL22) from keratinocytes [29], which attract Th2 cells through CC chemokine receptor 4 (CCR4) [30]. Serum TARC levels are known to correlate well with the severity of AD [31].

In humans, polymorphisms in the gene encoding the cytokine TSLP are associated with the development of multiple allergic disorders. It is known that TSLP is highly expressed in the epidermis of the lesional skin of AD patients [32], and that TSLP is induced by mechanical injury [33] and PAR-2 activation [13]. Overexpression of TSLP by keratinocytes results in an AD-like inflammatory phenotype in mice [34].

3.2. Dendritic cell (DC) subsets

Upon protein antigen exposure, DCs acquire antigens and stimulate the proliferation of T cells to induce distinct T helper cell responses to external pathogens [35]. Therefore, it has been suggested that DCs initiate AD in humans [36]; however, it remains unclear which cutaneous DC subset initiates epicutaneous sensitization to protein antigens. In mouse skin, there are at least three subsets of DCs [37–39]: Langerhans cells (LCs) in the epidermis and Langerin-positive and Langerin-negative DCs in the dermis (Langerin⁺ dermal DCs and Langerin[−] dermal DCs, respectively).

It has been reported that the application of large molecules is localized above the size-selective barrier, TJ, and that activated LCs extend their dendrites through the TJ to take up antigens [40] (Fig. 3). Therefore, it can be hypothesized that it is not dermal DCs but rather LCs that initiate epicutaneous sensitization with protein antigens, as in the development of AD.

It has recently been demonstrated that LCs are the essential cutaneous DC subset in the induction of IgE upon epicutaneous sensitization with the protein antigen, ovalbumin (OVA), that TSLP receptor expression on LCs is enhanced upon protein antigen exposure to the skin, and that LCs play an important role in this process through TSLP-TSLP receptor signaling [41]. Consistent with the above finding, in a mouse model of AD induced by a vitamin D3

Table 1
Characteristics of extrinsic and intrinsic AD [5].

	Extrinsic	Intrinsic
Frequency	80%	20%
Serum IgE	Elevated	Normal
Filaggrin mutation	High	Normal
Incidence (male:female)	1:1	1:3–4
Onset	Early	Relatively late
Skin barrier	Disrupted	Normal
Immunological feature	Th2 dominant	Th1 dominant
Metal allergy	Not related	High prevalence

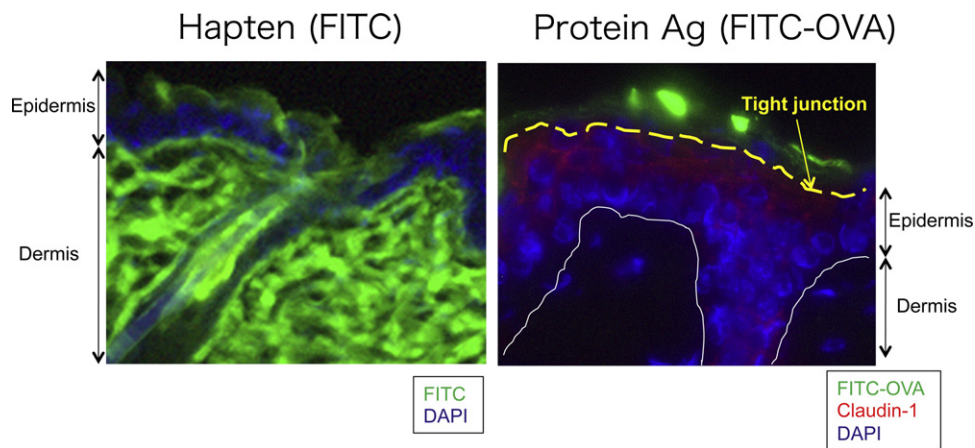


Fig. 3. Distribution of haptens and protein antigens upon epicutaneous application. After the epicutaneous application of green-fluorescent hapten (FITC) and protein antigen (FITC-ovalbumin; FITC-OVA), haptens penetrate the dermis, whereas protein antigens remained at the stratum corneum.

analog, epidermal LC-depleted mice treated with vitamin D3 did not exhibit AD-like inflammation or increased serum IgE compared to vitamin D3 analog-treated control mice [42]. Similar findings were also observed in a murine model of experimental staphylococcal scalded skin syndrome [43].

On the other hand, in a CHS model of hapten, LCs seem to be indispensable. Haptens can easily penetrate into the dermis upon topical application (Fig. 3). Langerin⁺ dermal DCs probably exert stimulatory effects during sensitization. The function of Langerin[−] dermal DCs in sensitization has not yet been investigated intensively; however, Langerin[−] dermal DCs also appear to have stimulatory functions in sensitization because ablation of both LCs and Langerin⁺ dermal DCs prior to sensitization impairs the CHS response but is unable to abrogate it completely [38,44,45]. This issue is discussed in more detail (Sections 3.4 and 3.5) below.

3.3. T cell subsets

3.3.1. Th1/Th2 paradigms

AD has been considered as an allergic Th2-mediated disease that is characterized by abnormal IgE production, peripheral eosinophilia, mast cell activation, and induction of Th2 lymphocytes expressing IL-4 and IL-13 [1] (Fig. 4).

On the other hand, sequential biopsies in AD patients after exposure to aeroallergens, such as house dust mites, in patch tests demonstrated a biphasic immunologic response characterized by a switch from a Th2 to a Th1 phenotype in later phases of the disease [46]. It remains unknown how the Th1 shift occurs in the clinical setting. The Th2 environment is known to inhibit the production of antimicrobial peptides in the skin, which predispose the skin to superimposed infection [47]. Therefore, one possible explanation for Th2 to Th1 shifting in the chronic phase of AD is that superimposed infection of *Staphylococcus aureus* shifts the skin conditions toward Th1. Intriguingly, the microbial colonization by *S. aureus* amplifies inflammation, and further stresses the skin barrier in AD.

3.3.2. Th17 and T22

In addition to the Th1/Th2 paradigm, the roles of Th17 and regulatory T cells (Treg) have been highlighted [48–51]. Transforming growth factor (TGF)- β , IL-1 β , IL-6, and IL-23 are key factors involved in naïve T cell commitment to a Th17 phenotype (Fig. 4). Th17 cells are characterized by the production of inflammatory cytokines, such as IL-17A, IL-17F, IL-22, and IL-26. Th17 cells have been implicated in several human autoimmune diseases, including multiple sclerosis, rheumatoid arthritis, inflammatory bowel disease, and psoriasis [52].

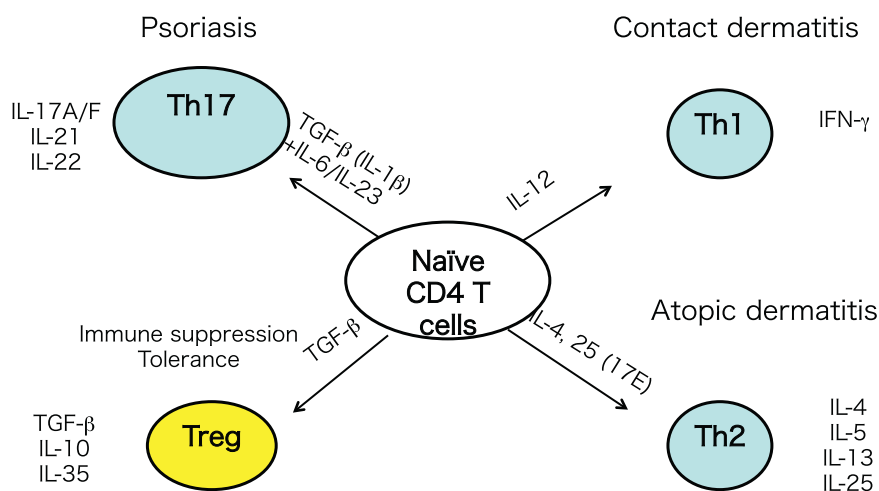


Fig. 4. T cells subsets. CD4⁺ helper T (Th) cells have at least three subtypes, Th1, Th2, and Th17, that are involved in the pathogenesis of contact dermatitis, AD/urticaria, and psoriasis, respectively. These Th subtypes are induced by specific cytokine conditions. On the other hand, regulatory T cells (Treg) are localized in the skin and play an important role in maintaining homeostasis and terminating a variety of immune responses in the skin.

Epicutaneous immunization in mice with OVA induces allergic skin inflammation resembling AD together with cutaneous expression of IL-17A and IL-17F mRNA, and, subsequently, airway inflammation [53]. In addition, the presence of Th17 cells and IL-17 in AD patients has been demonstrated [54,55]. Phenotypic analysis of peripheral blood mononuclear cells derived from AD patients exhibited a marked increase in the IL-17⁺CD4⁺ T-cell population. A high percentage of IL-17-producing cells was found in severe AD [54,55], which suggests a direct correlation between the presence of Th17 cells and disease severity. Increased levels of IL-17 were also reported in the cutaneous lesions of AD patients. Consistently, *Figt*^{fl} mice were shown to possess increased IL-17 mRNA levels in skin lesions shortly after the epicutaneous application of OVA [23]. It has been demonstrated that IL-17A and IL-17F promote eosinophil production of CXCL1, IL-8, and CCL4, as well as IL-1 β and IL-6, indicating that there is possible cross-talk between Th17 cells and eosinophils in AD [56]. In addition, histamine appears to enhance the effect of IL-17 on keratinocytes to produce inflammatory mediators in the presence of other pro-inflammatory cytokines, including TNF- α , through the histamine H1-receptor [57].

On the other hand, distinct IL-22-producing CD4⁺ and CD8⁺ T-cell populations (called T22) were significantly increased in AD skin. IL-22⁺CD8⁺ T-cell frequency correlated with AD disease severity [58]. Therefore, the above findings suggest that a Th2/T22 polarization dominates chronic AD. CCL27, the ligand for CCR10, is highly expressed in skin lesions from AD patients [59] and, combined with the recent finding that T22 cells particularly express CCR10, may be an explanation for the increased T22 presence in chronic AD skin. IL-22 has been shown to induce epidermal proliferation with acanthosis and hypogranulosis in skin through the downregulation of genes involved in terminal differentiation of the skin [60], which may be involved in the development of AD.

3.3.3. Regulatory T cells (Treg)

Treg exist in all non-lymphoid tissues and induce immune suppression or tolerance [49,50]. The skin has a high proportion of Treg in the steady state in mice and humans [51,61,62]. Treg in the skin are CD44⁺ and CD103^{high} [51,61,62], and they express the chemokine receptors, CCR4, CCR5, CCR6, and CCR7. Mice with a complete loss of CCR4 in Treg develop spontaneous lymphocytic infiltration and severe inflammation in the skin and lungs, accompanied by peripheral lymphadenopathy and increased differentiation of skin-tropic CD4⁺Foxp3⁺ memory T cells. Using α -1, 3-fucosyltransferase VII (Fut7)-deficient mice, Dudda et al. have reported the importance of E- and P-selectin ligands for Treg migration to the skin [62]. Loss of the bindings of selectins caused skin-specific inflammation, indicating the essential role of skin-resident Treg in maintaining immune homeostasis locally.

It has also been reported that Treg is accumulated in the skin during CHS [51] and that Treg suppress both the sensitization and elicitation of the CHS response [49,50,63]. In addition, IL-10 is induced in the repeat hapten application-induced chronic CHS model [64]. These findings demonstrate that chronic antigen exposure induces Treg accumulation in the skin (Fig. 5). Clinically, topical immunotherapy with squaric acid dibutylester (SADBE) is effective for the treatment of alopecia areata [65]. It remains unclear how SADBE controls this autoimmune disease, but the accumulation of Treg in the chronic hapten-exposed skin may play an important role (Fig. 5).

It is known that patients with immunodysregulation poly-endocrinopathy enteropathy X-linked (IPEX) syndrome demonstrate atopic-like dermatitis and high IgE levels as well as loss of Treg functions [66]. On the contrary, it has also been reported that the number of Treg in peripheral blood is similar between AD and

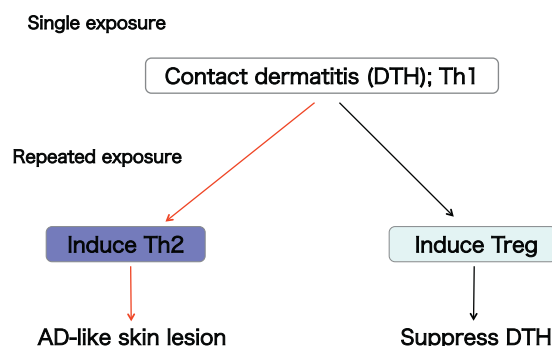


Fig. 5. Dynamics of cutaneous immune responses. A single hapten elicitation induces Th1-mediated delayed-type hypersensitivity (DTH), known as contact dermatitis/contact hypersensitivity. Even during the contact dermatitis response, Treg accumulates in the skin and suppresses DTH responses. Repeated hapten elicitation induces Th2 conditions in the skin, which is characteristic of AD.

healthy controls [67]. As for Treg in AD skin lesions, it was first reported that Treg were absent in the lesions of AD, although Tr1 was detected [68]. Later on, however, several groups reported the existence of Treg in the skin lesions of AD [69,70]. Reefer et al., on the other hand, reported that a new subtype of Treg with Th2-promoting ability was observed in AD and that the functions depend on the expression of CCR6 [71].

AD is a chronic inflammatory disease that has multiple disease stages. Treatment with drugs such as cyclosporine [67,72,73], glucocorticoid [74], and UV [75] can alter the number of Treg in the blood. Therefore, the interpretation and comparison of each study require careful attention.

3.4. Immune reactions to haptens

Haptens are external antigens that easily penetrate into the dermis (Fig. 3). As is well known, a single hapten application induces a classic delayed-type hypersensitivity, called the CHS response, which is mediated by IFN- γ -producing CD8⁺ and CD4⁺ T cells [35].

LCs have long been regarded as essential antigen-presenting cells for the establishment of sensitization in hapten-induced CHS, but this concept is now being challenged by recent analyses using LC ablation murine models [76]. In the development of CHS to haptens, Langerin-negative DCs play a major role, and LCs and Langerin-positive dermal DCs play a compensatory role [44].

On the other hand, it has been established that the repeated application of haptens (2,4,6-trinitrochlorobenzene; TNCB) induce AD-like skin lesions [77]. In this model, whilst a single challenge induced CHS, a prototype of delayed-type hypersensitivity, repeated hapten application results in a shift from Th1- to Th2-mediated cutaneous inflammation with elevated IL-4 expression, eosinophil infiltration in the skin, and elevated hapten-specific serum IgE levels [77]. In addition, extraordinary sustained IFN- γ expression leads to a Th1-dominant state in the mouse AD model with clinical amelioration of AD symptoms [78,79].

This concept is depicted as the “Hapten-Atopy hypothesis” [80,81]. It has been postulated that initial innate immune responses to chemical haptens result in the promotion of Th1 cell responses. On the other hand, under conditions where there is prolonged skin exposure to hapten, there will be a shift of Th cell phenotype to selective Th2-type responses [80]. Clinicians should be aware of increased levels of contact sensitization in individuals with AD [81]. At present, which cells mediate the shift from Th1 to Th2 has been a topic of debate. One of the candidates appears to include basophils, which express MHC class II and IL-4 in the draining lymph nodes (unpublished observation) (Fig. 6).

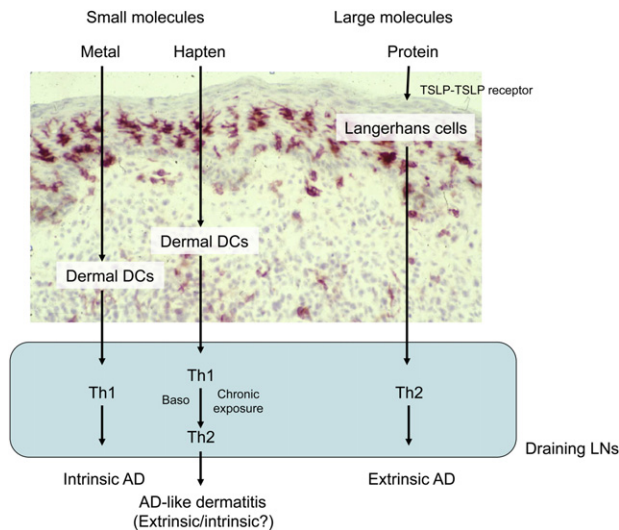


Fig. 6. Cutaneous immune responses to foreign antigens. Small molecules, such as metal and hapten, penetrate the dermis; therefore, dermal dendritic cells are responsible for the induction of immune responses. Metal exposure may lead to intrinsic AD without inducing Th2/IgE. Hapten exposure will initially provoke Th1, followed by Th2 after repeated exposure. On the other hand, protein antigens are captured by Langerhans cells that induce Th2 via TSLP-TSLP receptor signaling.

Metal is also known to be a contact sensitizer [82]. It remains unclear whether chronic metal exposure induces similar clinical manifestations as observed in AD, but it has been reported that intrinsic AD patients tend to exhibit positivity for metal patch tests without elevated IgE [5,28] (Fig. 6).

3.5. Immune reactions to protein antigens

Unlike haptens, conventional allergens for AD are rather large (Figs. 3 and 6). Therefore, LCs are thought to be the DCs responsible for acquiring cutaneous allergens, such as house dust mites, in the development of AD, as mentioned in Section 3.2.

It is known that the epicutaneous application of the protein antigen, OVA, induces a rise in OVA-specific serum IgE and IgG1, both of which are induced in a Th2-dependent manner, as well as

the development of dermatitis characterized by the infiltration of CD3⁺ T cells, eosinophils, and neutrophils and the local expression of mRNA for the cytokines IL-4, IL-5, and, intriguingly, IFN- γ [83]. Consistently, chronic exposure to protein antigens, especially those with protease activities (e.g., house dust mite allergens), induces TSLP expression in the epidermis. The above findings suggest that chronic exposure to hapten or protein antigen induce AD-like skin lesions in mice, which fits well with the pathogenesis of human AD (Fig. 6).

3.6. Interplay between barrier and allergy/immunology

It has been investigated whether acute removal of the stratum corneum modulates the production of cytokines and chemokines by epidermal cells. Tape stripping upregulates TSLP levels in the skin, which polarizes skin DCs to elicit a Th2 response [33]. Therefore, barrier disruption seems to deviate the skin environment toward Th2. In addition, Th2 chemokines (CCL17 and CCL22) and eosinophil chemoattractant (CCL5) mRNA levels were markedly elevated in mice by barrier disruption, which was augmented by tape stripping more markedly than by acetone rubbing [29]. In addition, tape stripping induced dermal infiltration of eosinophils in mice [29]. These findings suggest that acute barrier removal induces the expression of Th2 and eosinophil chemokines by epidermal cells and easily evokes the late phase reaction upon a challenge with antigen. These findings suggest that the barrier dysfunction predisposes the skin environment to Th2 skewing conditions as well as the feasibility for antigen exposure to the internal skin (Fig. 7). Therefore, under certain conditions, especially when the filaggrin is mutated or if scratching disrupts the skin barrier, the barrier dysfunction is the primary cause in the development of AD.

Intriguingly, on the other hand, human keratinocytes differentiated in the presence of IL-4 and IL-13 exhibited significantly reduced *FLG* gene expression [84] (Fig. 7). In addition, IL-17A downregulates the expression of filaggrin and genes that are important for cellular adhesion, which leads to impairment of epidermal barrier formation [85]. Consistently, the level of *FLG* expression in AD patients even without *FLG* mutations was decreased. These findings indicate that the atopic inflammatory response induces an acquired barrier defect to create a positive feedback loop through a highly complex interplay.

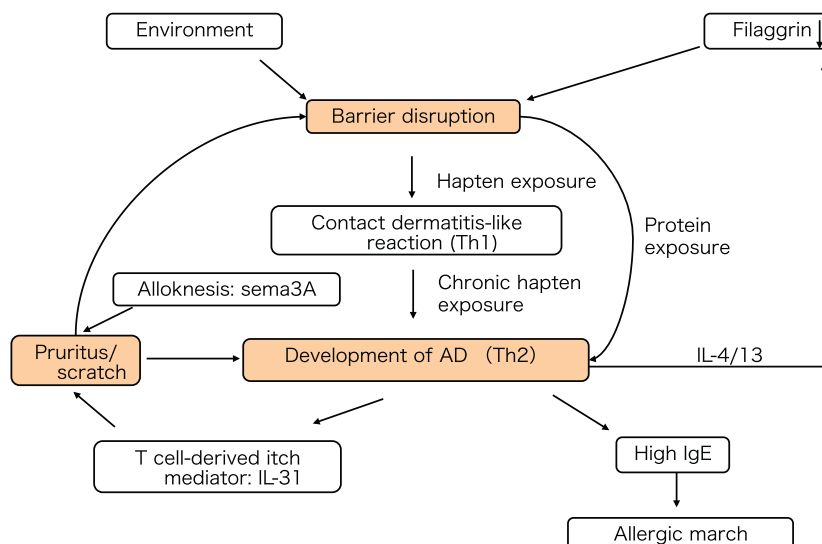


Fig. 7. Interplay among barrier, allergy/immunology, and pruritus. As shown in this figure, the development of AD involves a highly complex interplay among barrier disruption, allergy/immunology, and pruritus/scratching.

4. Pruritus in AD

4.1. Pruritus

Itching, or pruritus, is defined as an unpleasant cutaneous sensation that serves as a physiological self-protective mechanism to prevent the body from being hurt by harmful external agents. Chronic itch represents a significant clinical problem resulting from renal diseases and liver diseases, as well as several serious skin diseases, including AD. The gastrin-releasing peptide receptor or Mrgpr_s mediates itch sensation in the dorsal spinal cord/C-fiber [86,87].

Pruritus is known to be induced by a variety of mediators [88] (Table 2). Among them, histamine is a well-known pruritogen, since antihistamines (H1 blockers) are effective at controlling pruritus in several skin diseases, especially in urticaria. However, pruritus in AD is not controlled well by antihistamines, suggesting that there exist other itch mediators for it.

The sprouting of epidermal nerve fibers that is associated with pruritus is found in human and animal AD [89–91]. This condition explains the characteristic itchy skin (alloknesis) that is produced by innocuous mechanical stimulation in patients with AD. Nerve growth factor (NGF) produced by keratinocytes determines skin innervation. Increased plasma concentrations of NGF in AD patients are correlated with disease severity [92]. Therefore, epidermal innervation is regulated by the fine-tuning between nerve elongation factors (e.g., NGF, amphiregulin, gelatinase) and nerve repulsion factors (e.g., semaphorin [Sema]3A, anosmin-1) [93]. Topically applied Sema3A ointment is known to inhibit scratching behavior and improves skin inflammation in NC/Nga mice as a model of AD [94]. Intriguingly, histamine H4 receptor antagonist JNJ7777120 attenuated scratching behavior and improved dermatitis. These effects were augmented by combined treatment with H1 receptor antagonist olopatadine, which had a similar therapeutic efficacy to prednisolone. In addition, olopatadine reversed the histamine-induced reduction of Sema3A mRNA in keratinocytes [95].

Pruritus in AD is also regulated in the dermis. Histamine reduces Sema3A production by fibroblasts, allowing C-fibers to elongate in the dermis. In contrast, the histamine-augmented keratinocyte production of Sema3A may suppress C-fiber elongation and result in exaggerated pruritus [96]. It has recently been reported that dermal fibroblasts secrete artemin in response to substance P, leading to abnormal peripheral innervation and thermal hyperalgesia [97]. It is probable that artemin lowers the

threshold of temperature-dependent itch sensation in pruritic skin disorders, including AD.

4.2. Interplay between pruritus and barrier

The interplay between pruritus and the barrier is rather evident, since scratching directly disrupts skin barrier functions. On the other hand, once the barrier is disrupted, as a result of acetone application, for example, intraepidermal nerve fibers are known to increase in the skin in accordance with increased epidermal NGF and amphiregulin levels [91], which suggests that dry skin/barrier disruption predisposes the skin to pruritus. The acetone-induced intraepidermal nerve growth was significantly decreased by phototherapy, in accordance with the normalization of the abnormal expression of NGF and Sema3A in the epidermis [93].

4.3. Interplay between pruritus and allergy/immunology

Whether or not scratching affects immune functions has been an issue of debate. Recently, it was reported that tape stripping, which has a similar physiological effect to scratching, induced eosinophil infiltration into the skin in line with TARC/CCL27 and MDC/CCL22 induction in keratinocytes [29], which attract Th2 cells via CCR4 [30]. In addition, mechanical injury induces the expression of TSLP [33]. Therefore, pruritus/scratching leads to Th2 skewing conditions in the skin.

In terms of the effect of immune responses on pruritus, it is well known that cyclosporine suppresses pruritus in patients with AD much sooner than the skin lesions improve, suggesting the existence of T cell-mediated itch mediators. One of the candidates involved in pruritus is IL-31, which has been reported to be associated with pruritus in AD [98,99]. T cells (especially Th2 cells) from AD lesions contained significantly higher percentages of IL-31-producing T cells [98,99]. IL-31 receptor A is most abundantly expressed in the dorsal root ganglia representing the site where the cell bodies of cutaneous sensory neurons reside [98]. Consistently, systemic cyclosporine treatment suppressed the serum level of IL-31 [100]. These findings suggest that there is interplay between allergy/immunology and pruritus in AD, possibly via Th2-derived IL-31.

Familial primary localized cutaneous amyloidosis (FPLCA) is an autosomal-dominant disorder associated with chronic skin itching and deposition of epidermal keratin filament-associated amyloid material in the dermis. Recently, missense mutations in the OSMR gene, encoding oncostatin M-specific receptor beta, were detected in FPLCA [101]. Since OSMRbeta is a component of the oncostatin M type II receptor and the IL-31 receptor, the relationship between pruritus and IL-31 is of interest.

5. Future direction

In this article, the roles of three important elements in AD: barrier, allergy/immunology, and pruritus, were reviewed independently. The interplay between each factor was then discussed, as illustrated in Fig. 7. These interactions are important in understanding the pathomechanism of AD because they are linked to each other as a trinity. Therefore, in the development of AD, barrier dysfunction, allergy/immunological dysregulation, and pruritus create a positive feedback loop through a highly complex interplay.

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Table 2
Mediators in pruritus.

1. Peripheral mediators		
Source	Mediators	Receptors
Mast cells	Histamine	H1 (H4?)
Nerves	Substance P	
Eosinophils	Reactive oxygen	
Fibroblasts	Artemin	
Keratinocytes	Nerve growth factor, semaphorin 3A	
T cells	IL-2	IL-2 receptor
	IL-31	IL-31 receptor
Endothelial cells	Endothelin	
	Acetylcholine	ACh receptor
2. Central mediators		
Mediators	Receptors	
Enkephalin		
Morphine/opioid	μ receptor (pruritogenic) κ receptor (anti-pruritogenic)	

References

- [1] Bieber T. Atopic dermatitis. *N Engl J Med* 2008;358:1483–94.
- [2] Akdis CA, Akdis M, Bieber T, Bindslev-Jensen C, Boguniewicz M, Eigenmann P, et al. Diagnosis and treatment of atopic dermatitis in children and adults: European Academy of Allergy and Clinical Immunology/American Academy of Allergy, Asthma and Immunology/PRACTALL Consensus Report. *J Allergy Clin Immunol* 2006;118:152–69.
- [3] Heilmann J, Spengel JM. Filaggrin mutations and atopy: consequences for future therapeutics. *Exp Rev Clin Immunol* 2012;8:189–97.
- [4] Egawa G, Kabashima K. Skin as a peripheral lymphoid organ: revisiting the concept of skin-associated lymphoid tissues. *J Invest Dermatol* 2011;131:2178–85.
- [5] Tokura Y. Extrinsic and intrinsic types of atopic dermatitis. *J Dermatol Sci* 2010;58:1–7.
- [6] Johansson O, Liang Y, Heilborn JD, Marcusson JA. Langerhans cells in prurigo nodularis investigated by HLA-DR and S-100 immunofluorescence double staining. *J Dermatol Sci* 1998;17:24–32.
- [7] Kabashima K. Pathomechanism of atopic dermatitis in the perspective of T cell subsets and skin barrier functions – “Which comes first, the chicken or the egg?” *Dermatol Sin* 2012;30:142–6.
- [8] Kaplan DH, Igyarto BZ, Gaspari AA. Early immune events in the induction of allergic contact dermatitis. *Nat Rev Immunol* 2012;12:114–24.
- [9] Rippke F, Schreiner V, Doering T, Maibach HI. Stratum corneum pH in atopic dermatitis: impact on skin barrier function and colonization with *Staphylococcus aureus*. *Am J Clin Dermatol* 2004;5:217–23.
- [10] Briot A, Deraison C, Lacroix M, Bonnart C, Robin A, Besson C, et al. Kallikrein 5 induces atopic dermatitis-like lesions through PAR2-mediated thymic stromal lymphopoietin expression in Netherton syndrome. *J Exp Med* 2009;206:1135–47.
- [11] Palmer CN, Irvine AD, Terron-Kwiatkowski A, Zhao Y, Liao H, Lee SP, et al. Common loss-of-function variants of the epidermal barrier protein filaggrin are a major predisposing factor for atopic dermatitis. *Nat Genet* 2006;38:441–6.
- [12] Moniaga CS, Egawa G, Kawasaki H, Hara-Chikuma M, Honda T, Tanizaki H, et al. Flaky tail mouse denotes human atopic dermatitis in the steady state and by topical application with *Dermatophagoides pteronyssinus* extract. *Am J Pathol* 2010;176:2385–93.
- [13] Moniaga CS, Jeong SK, Egawa G, Nakajima S, Hara-Chikuma M, Jeon JE, et al. Protease activity enhances production of thymic stromal lymphopoietin and basophil accumulation in flaky tail mice. *Am J Pathol*, PMID: 23333753, in press.
- [14] Furuse M, Hata M, Furuse K, Yoshida Y, Haratake A, Sugitani Y, et al. Claudin-based tight junctions are crucial for the mammalian epidermal barrier: a lesson from claudin-1-deficient mice. *J Cell Biol* 2002;156:1099–111.
- [15] Hadj-Rabia S, Baala L, Vabres P, Hamel-Teillac D, Jacquemin E, Fabre M, et al. Claudin-1 gene mutations in neonatal sclerosing cholangitis associated with ichthyosis: a tight junction disease. *Gastroenterology* 2004;127:1386–90.
- [16] Vijayanand P, Seumois C, Simpson LJ, Abdul-Wajid S, Baumjohann D, Panduro M, et al. Interleukin-4 production by follicular helper T cells requires the conserved IL4 enhancer hypersensitivity site V. *Immunity* 2012;36(2):175–87.
- [17] Proksch E, Folster-Holst R, Jensen JM. Skin barrier function, epidermal proliferation and differentiation in eczema. *J Dermatol Sci* 2006;43:159–69.
- [18] Candi E, Schmidt R, Melino G. The cornified envelope: a model of cell death in the skin. *Nat Rev Mol Cell Biol* 2005;6:328–40.
- [19] Listwan P, Rothnagel JA. Keratin bundling proteins. *Methods Cell Biol* 2004;78:817–27.
- [20] Brown SJ, Kroboth K, Sandilands A, Campbell LE, Pohler E, Kezic S, et al. Intragenic copy number variation within filaggrin contributes to the risk of atopic dermatitis with a dose-dependent effect. *J Invest Dermatol* 2012;132:98–104.
- [21] Moniaga CS, Kabashima K. Filaggrin in atopic dermatitis: flaky tail mice as a novel model for developing drug targets in atopic dermatitis. *Inflamm Allergy Drug Targets* 2011;10:477–85.
- [22] Fallon PG, Sasaki T, Sandilands A, Campbell LE, Saunders SP, Mangan NE, et al. A homozygous frameshift mutation in the mouse Flg gene facilitates enhanced percutaneous allergen priming. *Nat Genet* 2009;41:602–8.
- [23] Oyoshi MK, Murphy GF, Geha RS. Filaggrin-deficient mice exhibit TH17-dominated skin inflammation and permissiveness to epicutaneous sensitization with protein antigen. *J Allergy Clin Immunol* 2009;124:485–93. 493.e1.
- [24] Brown SJ, Asai Y, Cordell HJ, Campbell LE, Zhao Y, Liao H, et al. Loss-of-function variants in the filaggrin gene are a significant risk factor for peanut allergy. *J Allergy Clin Immunol* 2011;127:661–7.
- [25] Kawasaki H, Nagao K, Kubo A, Hata T, Shimizu A, Mizuno H, et al. Altered stratum corneum barrier and enhanced percutaneous immune responses in filaggrin-null mice. *J Allergy Clin Immunol* 2012;129:1538–46. e6.
- [26] Mori T, Ishida K, Mukumoto S, Yamada Y, Imokawa G, Kabashima K, et al. Comparison of skin barrier function and sensory nerve electric current perception threshold between IgE-high extrinsic and IgE-normal intrinsic types of atopic dermatitis. *Br J Dermatol* 2010;162:83–90.
- [27] Kabashima-Kubo R, Nakamura M, Sakabe J, Sugita K, Hino R, Mori T, et al. A group of atopic dermatitis without IgE elevation or barrier impairment shows a high Th1 frequency: possible immunological state of the intrinsic type. *J Dermatol Sci* 2012;67:37–43.
- [28] Shanon J. Pseudo-atopic dermatitis. Contact dermatitis due to chrome sensitivity simulating atopic dermatitis. *Dermatologica* 1965;131:176–90.
- [29] Onoue A, Kabashima K, Kobayashi M, Mori T, Tokura Y. Induction of eosinophil- and Th2-attracting epidermal chemokines and cutaneous late-phase reaction in tape-stripped skin. *Exp Dermatol* 2009;18:1036–43.
- [30] Nickel R, Beck LA, Stellato C, Schleimer RP. Chemokines and allergic disease. *J Allergy Clin Immunol* 1999;104:723–42.
- [31] Shimada Y, Takehara K, Sato S. Both Th2 and Th1 chemokines (TARC/CCL17, MDC/CCL22, and Mig/CXCL9) are elevated in sera from patients with atopic dermatitis. *J Dermatol Sci* 2004;34:201–8.
- [32] Soumelis V, Reche PA, Kanzler H, Yuan W, Edward G, Homey B, et al. Human epithelial cells trigger dendritic cell mediated allergic inflammation by producing TSLP. *Nat Immunol* 2002;3:673–80.
- [33] Oyoshi MK, Larson RP, Ziegler SF, Geha RS. Mechanical injury polarizes skin dendritic cells to elicit a T(H)2 response by inducing cutaneous thymic stromal lymphopoietin expression. *J Allergy Clin Immunol* 2010;126:976–84. 984.e1–5.
- [34] Yoo J, Omori M, Gyarmati D, Zhou B, Aye T, Brewer A, et al. Spontaneous atopic dermatitis in mice expressing an inducible thymic stromal lymphopoietin transgene specifically in the skin. *J Exp Med* 2005;202:541–9.
- [35] Honda T, Egawa G, Grabbe S, Kabashima K. Update of immune events in the murine contact hypersensitivity model: toward the understanding of allergic contact dermatitis. *J Invest Dermatol* 2013;133:303–15.
- [36] Novak N, Peng W, Yu C. Network of myeloid and plasmacytoid dendritic cells in atopic dermatitis. *Adv Exp Med Biol* 2007;601:97–104.
- [37] Ginhoux F, Collin MP, Bogunovic M, Abel M, Leboeuf M, Helft J, et al. Blood-derived dermal Langerin + dendritic cells survey the skin in the steady state. *J Exp Med* 2007;204:3133–46.
- [38] Bursch LS, Wang L, Igyarto B, Kissenpfennig A, Malissen B, Kaplan DH, et al. Identification of a novel population of Langerin + dendritic cells. *J Exp Med* 2007;204:3147–56.
- [39] Poulin LF, Henri S, de Bovis B, Devillard E, Kissenpfennig A, Malissen B. The dermis contains Langerin + dendritic cells that develop and function independently of epidermal Langerhans cells. *J Exp Med* 2007;204:3119–31.
- [40] Kubo A, Nagao K, Yokouchi M, Sasaki H, Amagai M. External antigen uptake by Langerhans cells with reorganization of epidermal tight junction barriers. *J Exp Med* 2009;206:2937–46.
- [41] Nakajima S, Igyarto BZ, Honda T, Egawa G, Otsuka A, Hara-Chikuma M, et al. Langerhans cells are critical in epicutaneous sensitization with protein antigen via thymic stromal lymphopoietin receptor signaling. *J Allergy Clin Immunol* 2012;129:1048–55. e6.
- [42] Elentner A, Finke D, Schmuth M, Chappaz S, Ebner S, Malissen B, et al. Langerhans cells are critical in the development of atopic dermatitis-like inflammation and symptoms in mice. *J Cell Mol Med* 2009;13:2658–72.
- [43] Ouchi T, Kubo A, Yokouchi M, Adachi T, Kobayashi T, Kitashima DY, et al. Langerhans cell antigen capture through tight junctions confers preemptive immunity in experimental staphylococcal scalded skin syndrome. *J Exp Med* 2011;208:2607–13.
- [44] Honda T, Nakajima S, Egawa G, Ogasawara K, Malissen B, Miyachi Y, et al. Compensatory role of Langerhans cells and Langerin-positive dermal dendritic cells in the sensitization phase of murine contact hypersensitivity. *J Allergy Clin Immunol* 2010;125:1154–6. e2.
- [45] Wang L, Bursch LS, Kissenpfennig A, Malissen B, Jameson SC, Hogquist KA. Langerin expressing cells promote skin immune responses under defined conditions. *J Immunol* 2008;180:4722–7.
- [46] Grewe M, Walther S, Gyufko K, Czech W, Schopf E, Krutmann J. Analysis of the cytokine pattern expressed in situ in inhalant allergen patch test reactions of atopic dermatitis patients. *J Invest Dermatol* 1995;105:407–10.
- [47] Nomura I, Goleva E, Howell MD, Hamid QA, Ong PY, Hall CF, et al. Cytokine milieu of atopic dermatitis, as compared to psoriasis, skin prevents induction of innate immune response genes. *J Immunol* 2003;171:3262–9.
- [48] Di Cesare A, Di Meglio P, Nestle FO. A role for Th17 cells in the immunopathogenesis of atopic dermatitis? *J Invest Dermatol* 2008;128:2569–71.
- [49] Honda T, Miyachi Y, Kabashima K. The role of regulatory T cells in contact hypersensitivity. *Recent Pat Inflamm Allergy Drug Discov* 2010;4:85–9.
- [50] Honda T, Miyachi Y, Kabashima K. Regulatory T cells in cutaneous immune responses. *J Dermatol Sci* 2011;63:75–82.
- [51] Tomura M, Honda T, Tanizaki H, Otsuka A, Egawa G, Tokura Y, et al. Activated regulatory T cells are the major T cell type emigrating from the skin during a cutaneous immune response in mice. *J Clin Invest* 2010;120:883–93.
- [52] Tesmer LA, Lundy SK, Sarkar S, Fox DA. Th17 cells in human disease. *Immunol Rev* 2008;223:87–113.
- [53] He R, Oyoshi MK, Jin H, Geha RS. Epicutaneous antigen exposure induces a Th17 response that drives airway inflammation after inhalation challenge. *Proc Natl Acad Sci USA* 2007;104:15817–22.
- [54] Toda M, Leung DY, Molet S, Boguniewicz M, Taha R, Christodoulopoulos P, et al. Polarized in vivo expression of IL-11 and IL-17 between acute and chronic skin lesions. *J Allergy Clin Immunol* 2003;111:875–81.
- [55] Koga C, Kabashima K, Shiraishi N, Kobayashi M, Tokura Y. Possible pathogenic role of Th17 cells for atopic dermatitis. *J Invest Dermatol* 2008;128:2625–30.
- [56] Cheung PF, Wong CK, Lam CW. Molecular mechanisms of cytokine and chemokine release from eosinophils activated by IL-17A, IL-17F, and IL-23: implication for Th17 lymphocytes-mediated allergic inflammation. *J Immunol* 2008;180:5625–35.

- [57] Moniaga CS, Egawa G, Doi H, Miyachi Y, Kabashima K. Histamine modulates the responsiveness of keratinocytes to IL-17 and TNF- α through the H1-receptor. *J Dermatol Sci* 2011;61:79–81.
- [58] Nograles KE, Zaba LC, Shemer A, Fuentes-Duculan J, Cardinale I, Kikuchi T, et al. IL-22-producing “T22” T cells account for upregulated IL-22 in atopic dermatitis despite reduced IL-17-producing TH17 T cells. *J Allergy Clin Immunol* 2009;123:1244–52. e2.
- [59] Homey B, Alenius H, Muller A, Soto H, Bowman EP, Yuan W, et al. CCL27-CCR10 interactions regulate T cell-mediated skin inflammation. *Nat Med* 2002;8:157–65.
- [60] Boniface K, Bernard FX, Garcia M, Gurney AL, Lecron JC, Morel F. IL-22 inhibits epidermal differentiation and induces proinflammatory gene expression and migration of human keratinocytes. *J Immunol* 2005;174:3695–702.
- [61] Sather BD, Treuting P, Perdue N, Mizogowicz M, Fontenot JD, Rudensky AY, et al. Altering the distribution of Foxp3(+) regulatory T cells results in tissue-specific inflammatory disease. *J Exp Med* 2007;204:1335–47.
- [62] Dudda JC, Perdue N, Bachtanian E, Campbell DJ. Foxp3+ regulatory T cells maintain immune homeostasis in the skin. *J Exp Med* 2008;205:1559–65.
- [63] Honda T, Otsuka A, Tanizaki H, Minegaki Y, Nagao K, Waldmann H, et al. Enhanced murine contact hypersensitivity by depletion of endogenous regulatory T cells in the sensitization phase. *J Dermatol Sci* 2011;61:144–7.
- [64] Kitagaki H, Ono N, Hayakawa K, Kitazawa T, Watanabe K, Shiohara T. Repeated elicitation of contact hypersensitivity induces a shift in cutaneous cytokine milieu from a T helper cell type 1 to a T helper cell type 2 profile. *J Immunol* 1997;159:2484–91.
- [65] Rokhsar CK, Shupack JL, Vafai JJ, Washenik K. Efficacy of topical sensitizers in the treatment of alopecia areata. *J Am Acad Dermatol* 1998;39:751–61.
- [66] Ochs HD, Ziegler SF, Torgerson TR. FOXP3 acts as a rheostat of the immune response. *Immunol Rev* 2005;203:156–64.
- [67] Brandt C, Pavlovic V, Radbruch A, Worm M, Baumgrass R. Low-dose cyclosporine A therapy increases the regulatory T cell population in patients with atopic dermatitis. *Allergy* 2009;64:1588–96.
- [68] Verhagen J, Akdis M, Traidl-Hoffmann C, Schmid-Grendelmeier P, Hijnen D, Knol EF, et al. Absence of T-regulatory cell expression and function in atopic dermatitis skin. *J Allergy Clin Immunol* 2006;117:176–83.
- [69] Schnopp C, Rad R, Weidinger A, Weidinger S, Ring J, Eberlein B, et al. Fox-P3-positive regulatory T cells are present in the skin of generalized atopic eczema patients and are not particularly affected by medium-dose UVA1 therapy. *Photodermatol Photoimmunol Photomed* 2007;23:81–5.
- [70] Caproni M, Torchia D, Antiga E, Volpi W, del Bianco E, Fabbri P. The effects of tacrolimus ointment on regulatory T lymphocytes in atopic dermatitis. *J Clin Immunol* 2006;26:370–5.
- [71] Reefer AJ, Satinover SM, Solga MD, Lannigan JA, Nguyen JT, Wilson BB, et al. Analysis of CD25hiCD4+ “regulatory” T-cell subtypes in atopic dermatitis reveals a novel T(H)2-like population. *J Allergy Clin Immunol* 2008;121:415–22. e3.
- [72] Hijnen D, Haack I, van Kraats AA, Nijhuis E, de Bruin-Weller MS, Bruijnzeel-Koomen CA, et al. Cyclosporin A reduces CD4(+)CD25(+) regulatory T-cell numbers in patients with atopic dermatitis. *J Allergy Clin Immunol* 2009;124:856–8.
- [73] Baumgrass R, Brandt C, Wegner F, Abdollahnia M, Worm M. Low-dose, but not high-dose, cyclosporin A promotes regulatory T-cell induction, expansion, or both. *J Allergy Clin Immunol* 2010;126:183–4. author reply 184.
- [74] Stary G, Klein I, Bauer W, Koszik F, Reininger B, Kohlhofer S, et al. Glucocorticosteroids modify Langerhans cells to produce TGF- β and expand regulatory T cells. *J Immunol* 2011;186:103–12.
- [75] Loser K, Beissert S. Regulation of cutaneous immunity by the environment: an important role for UV irradiation and vitamin D. *Int Immunopharmacol* 2009;9:587–9.
- [76] Kissenpffennig A, Henri S, Dubois B, Laplace-Builhe C, Perrin P, Romani N, et al. Dynamics and function of Langerhans cells in vivo: dermal dendritic cells colonize lymph node areas distinct from slower migrating Langerhans cells. *Immunity* 2005;22:643–54.
- [77] Kitagaki H, Fujisawa S, Watanabe K, Hayakawa K, Shiohara T. Immediate-type hypersensitivity response followed by a late reaction is induced by repeated epicutaneous application of contact sensitizing agents in mice. *J Invest Dermatol* 1995;105:749–55.
- [78] Hattori K, Nishikawa M, Watcharanurak K, Ikoma A, Kabashima K, Toyota H, et al. Sustained exogenous expression of therapeutic levels of IFN- γ ameliorates atopic dermatitis in NC/Nga mice via Th1 polarization. *J Immunol* 2010;184:2729–35.
- [79] Mitsuishi T, Kabashima K, Tanizaki H, Ohsawa I, Oda F, Yamada Y, et al. Specific substance of Maruyama (SSM) suppresses immune responses in atopic dermatitis-like skin lesions in DS-Nh mice by modulating dendritic cell functions. *J Dermatol Sci* 2011;63:184–90.
- [80] McFadden JP, Dearman RJ, White JM, Basketter DA, Kimber I. The Hapten-Atopy hypothesis II: the ‘cutaneous hapten paradox’. *Clin Exp Allergy* 2011;41:327–37.
- [81] Thyssen JP, Linneberg A, Engkilde K, Menne T, Johansen JD. Contact sensitization to common haptens is associated with atopic dermatitis: new insight. *Br J Dermatol* 2012;166:1255–61.
- [82] Schmidt M, Raghavan B, Muller V, Vogl T, Fejer G, Tchapchet S, et al. Crucial role for human Toll-like receptor 4 in the development of contact allergy to nickel. *Nat Immunol* 2010;11:814–9.
- [83] Spergel JM, Mizoguchi E, Brewer JP, Martin TR, Bhan AK, Geha RS. Epicutaneous sensitization with protein antigen induces localized allergic dermatitis and hyperresponsiveness to methacholine after single exposure to aerosolized antigen in mice. *J Clin Invest* 1998;101:1614–22.
- [84] Howell MD, Kim BE, Gao P, Grant AV, Boguniewicz M, De Benedetto A, et al. Cytokine modulation of atopic dermatitis filaggrin skin expression. *J Allergy Clin Immunol* 2007;120:150–5.
- [85] Gutowska-Owsiak D, Schaupp AL, Salimi M, Selvakumar TA, McPherson T, Taylor S, et al. IL-17 downregulates filaggrin and affects keratinocyte expression of genes associated with cellular adhesion. *Exp Dermatol* 2012;21:104–10.
- [86] Sun YG, Chen ZF. A gastrin-releasing peptide receptor mediates the itch sensation in the spinal cord. *Nature* 2007;448:700–3.
- [87] Liu Q, Tang Z, Surdenikova L, Kim S, Patel KN, Kim A, et al. Sensory neuron-specific GPCR Mrgpr is itch receptors mediating chloroquine-induced pruritus. *Cell* 2009;139:1353–65.
- [88] Stander S, Steinhoff M. Pathophysiology of pruritus in atopic dermatitis: an overview. *Exp Dermatol* 2002;11:12–24.
- [89] Urashima R, Mihara M. Cutaneous nerves in atopic dermatitis. A histological, immunohistochemical and electron microscopic study. *Virchows Arch* 1998;432:363–70.
- [90] Tominaga M, Ozawa S, Ogawa H, Takamori K. A hypothetical mechanism of intraepidermal neurite formation in NC/Nga mice with atopic dermatitis. *J Dermatol Sci* 2007;46:199–210.
- [91] Tominaga M, Ozawa S, Tengara S, Ogawa H, Takamori K. Intraepidermal nerve fibers increase in dry skin of acetone-treated mice. *J Dermatol Sci* 2007;48:103–11.
- [92] Tominaga M, Ogawa H, Takamori K. Decreased production of semaphorin 3A in the lesional skin of atopic dermatitis. *Br J Dermatol* 2008;158:842–4.
- [93] Kamo A, Tominaga M, Tengara S, Ogawa H, Takamori K. Inhibitory effects of UV-based therapy on dry skin-inducible nerve growth in acetone-treated mice. *J Dermatol Sci* 2011;62:91–7.
- [94] Negi O, Tominaga M, Tengara S, Kamo A, Taneda K, Suga Y, et al. Topically applied semaphorin 3A ointment inhibits scratching behavior and improves skin inflammation in NC/Nga mice with atopic dermatitis. *J Dermatol Sci* 2012;66:37–43.
- [95] Ohsawa Y, Hirasawa N. The antagonism of histamine H1 and H4 receptors ameliorates chronic allergic dermatitis via anti-pruritic and anti-inflammatory effects in NC/Nga mice. *Allergy* 2012;67:1014–22.
- [96] Fukamachi S, Bito T, Shiraishi N, Kobayashi M, Kabashima K, Nakamura M, et al. Modulation of semaphorin 3A expression by calcium concentration and histamine in human keratinocytes and fibroblasts. *J Dermatol Sci* 2011;61:118–23.
- [97] Murota H, Izumi M, Abd El-Latif MI, Nishioka M, Terao M, Tani M, et al. Artemin causes hypersensitivity to warm sensation, mimicking warmth-provoked pruritus in atopic dermatitis. *J Allergy Clin Immunol* 2012;130:671–82. e4.
- [98] Sonkoly E, Muller A, Lauerma AI, Pivarcsi A, Soto H, Kemeny L, et al. IL-31: a new link between T cells and pruritus in atopic skin inflammation. *J Allergy Clin Immunol* 2006;117:411–7.
- [99] Dillon SR, Sprecher C, Hammond A, Bilsborough J, Rosenfeld-Franklin M, Presnell SR, et al. Interleukin 31, a cytokine produced by activated T cells, induces dermatitis in mice. *Nat Immunol* 2004;5:752–60.
- [100] Otsuka A, Tanioka M, Nakagawa Y, Honda T, Ikoma A, Miyachi Y, et al. Effects of cyclosporine on pruritus and serum IL-31 levels in patients with atopic dermatitis. *Eur J Dermatol* 2011;21:816–7.
- [101] Arita K, South AP, Hans-Filho G, Sakuma TH, Lai-Cheong J, Clements S, et al. Oncostatin M receptor-beta mutations underlie familial primary localized cutaneous amyloidosis. *Am J Hum Genet* 2008;82:73–80.



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