Pathophysiology of atopic dermatitis: Clinical implications

Jihyun Kim, M.D., Ph.D.,1–3 Byung Eui Kim, M.D., Ph.D.,2 and Donald Y. M. Leung, M.D., Ph.D.2

ABSTRACT

Atopic dermatitis (AD) is the most common chronic inflammatory skin disease. Genetic predisposition, epidermal barrier disruption, and dysregulation of the immune system are some of the critical components of AD. An impaired skin barrier may be the initial step in the development of the atopic march as well as AD, which leads to further skin inflammation and allergic sensitization. Type 2 cytokines as well as interleukin 17 and interleukin 22 contribute to skin barrier dysfunction and the development of AD. New insights into the pathophysiology of AD have focused on epidermal lipid profiles, neuroimmune interactions, and microbial dysbiosis. Newer therapeutic strategies focus on improving skin barrier function and targeting polarized immune pathways found in AD. Further understanding of AD pathophysiology will allow us to achieve a more precision medicine approach to the prevention and the treatment of AD.


Atopic dermatitis (AD) is the most common chronic inflammatory skin disease.1 The U.S. prevalence of AD was reported to be 11.3–12.7% and 6.9–7.6% in children and in adults, respectively.2 The Hanifin and Rajka criteria and the American Academy of Dermatology Consensus Criteria are useful diagnostic tools based on features of AD.3,4 AD severity can be assessed by using validated methods such as Scoring Atopic Dermatitis or the Eczema Area and Severity Index.5 Although the pathophysiology of AD is not completely understood, numerous studies demonstrated that skin barrier dysfunction and immune dysregulation contribute to the pathobiology of AD.6–8 The epidermis plays a crucial role as a physical and functional barrier, and skin barrier defects are the most significant pathologic findings in AD skin.9,10 Filaggrin (FLG), transglutaminases, keratins, and intercellular proteins are key proteins responsible for epidermal function. Defects in these proteins facilitate allergen and microbial penetration into the skin.11–13

Skin barrier dysfunction has been considered to be the first step in the development of atopic march as well as AD.7,12 However, it is also evident that immune dysregulation, including the activation of type 2 immune responses, results in impairment of the epidermal barrier.13–16 Recently, new insights into the pathophysiology of the development of AD focused on an important role of abnormalities in epidermal lipid layer as well as neuroimmune interactions and microbial dysbiosis.17–20 These factors have been used to develop novel therapeutic and preventative strategies of AD. This review addressed recent insights into the pathophysiologic mechanism of AD and the clinical application of these factors for improved treatment and prevention of AD. This work was supported by National Institutes of Health (grant AR41256). J. Kim and B. Eui Kim contributed equally to the article.

GENETICS

The filaggrin (FLG) gene is located on chromosome 1q2, and encodes FLG (filaggrin protein), which is a major structural protein in the stratum corneum (SC).21 Pro-FLG polymers are proteolytically cleaved and dephosphorylated into FLG monomers, which are associated with the aggregation of keratin filaments and the formation of SC.13 The generation of FLG degradation products, urocanic acid and pyrrolidine carboxylic acid, contributes to SC hydration and acidic pH of skin.14 It is well known that FLG null mutations impair skin barrier function and increase the risk of AD.21,22 FLG mutations, particularly homozygous mutations, are associated with an increased risk of severe AD with earlier onset, longer persistence, and skin infections.8,22,23 Approximately 10% of European populations are heterozygous carriers of FLG mutations, which results in a 50% reduction in expressed protein.22 However, the pathophysiology of AD goes far beyond FLG mutations. For example, Japanese and Korean patients have a lower frequency of FLG mutations than do patients in Western populations.13,24 Furthermore, ~40% of subjects with FLG-null alleles do not show characteristics of AD, and most of the pa-
patients with AD and with FLG mutations eventually outgrow the disease.25

Polymorphisms of various immune pathway genes are associated with an increased risk of AD through alternations in the T-helper (Th) type 2 signaling pathway.21,26 Upregulation of interleukin (IL) 4 and IL-13 lowers FLG expression, which leads to skin barrier defects.27,28 A gain of functional polymorphisms of type 2 cytokine receptors (IL-4R and IL-13R) are also implicated in AD pathogenesis.28,29 Other immune-related genes that contribute to the development of AD include IL-31, IL-33, signal transducer and activator of transcription (STAT) 6, thymic stromal lymphopoietin (TSLP) and its receptors (IL-7R and TSLPR), interferon regulatory factor 2, Toll-like receptor 2, and high-affinity IgE receptor (FceRI) α gene in specific populations.21,26,30–33 Additionally, recent studies demonstrated that vitamin D receptor polymorphisms and cytochrome P450 family 27 subfamily A member 1 (CYP27A1) variant are associated with AD.34,35 CYP27A1 is known to be involved in the metabolism of vitamin D3, which plays an essential role in immune modulation.34

Epigenetic mechanisms are heritable and can regulate gene expression without changing the DNA sequence.13 There is increasing evidence that demonstrates that environmental exposures induce epigenetic changes and AD through DNA modification and micro-RNA-mediated posttranscriptional regulation.26,36 A recent study provided evidence for the importance of DNA methylation and showed the relationship between umbilical cord blood methylation at 5′-C-phosphate-G-3′ sites of IL-4R and the development of AD at 1 year of age.37 DNA methylation in one adjacent CpG site of FLG was reported to have a significant interaction with FLG sequence variants and association with the increased risk of eczema.38 whereas another study, which used buccal cells, could not show the relationship between methylation of the FLG promoter and gene expression and allergic diseases.39 Furthermore, hypomethylation of TSLP and FceRI γ promoters contributes to gene overexpression in patients with AD.26

**IMMUNE DYSREGULATION**

Previous studies showed that type 2 immune cytokines, e.g., IL-4 and IL-13, play important roles in chemokine production, skin barrier dysfunction, suppression of antimicrobial peptides (AMP), and allergic inflammation.19,40 Interestingly, IL-31 was reported to enhance the release and production of brain-derived natriuretic peptide and to coordinate cytokine and chemokine release from skin cells, thereby inducing itch in patients with AD.41 In addition, TSLP is highly expressed in the epidermis of patients with AD, and its production is triggered by exposure to environmental factors such as allergens, microorganisms, diesel exhaust, cigarette smoke, and chemical irritants.13,42,43 When using skin tape samples, a Korean birth cohort study showed elevated expression of TSLP in the skin of 2 month-old infants before the development of clinical AD at 24 months of age.44

Although blockade of type 2–driven inflammation improves AD symptoms, the pathogenesis of AD is not exclusively explained by Th2 immunity. In this regard, IL-17 has been reported to reduce expression of FLG and involucrin.45,46 More prominent Th17 activation was observed in blood and acute AD skin lesions in Asian patients compared with European-American patients.47 In addition, AD is classified as the extrinsic and the intrinsic type, and production of IL-17 cytokine is higher in intrinsic AD with normal immunoglobulin E levels than in extrinsic AD.48 IL-22 is also highly upregulated in the skin of patients with AD and is associated with skin barrier dysfunction and abnormal epidermal markers, such as keratin 6 and keratin 16.49–51 In particular, transition to the chronic phase is manifested by the start of Th1-cell activation as well as the sustained activation of Th2 and Th22 cells (Fig. 1).52,53 Of interest, tumor necrosis factor α in combination with Th2 cytokines altered the expression of early and terminal differentiation products and reduced the level of long-chain free fatty acids (FFA) and ester linked ω-hydroxy (EO) ceramides.17,20

Recent studies showed that skin-resident group 2 innate lymphoid cells (ILC2) play a role in the pathogenesis of AD. ILC2s were found to produce IL-5 and IL-13, which result in the development of an AD-like skin lesion.54,55 Similarly, human skin ILC2s are highly enriched in lesional skin of patients with AD and activated by the epithelial cell–derived cytokines such as IL-25, IL-33, and/or TSLP.55,56 This leads to the production of type 2 cytokines and skin allergic inflammation.55,56 In contrast, epidermal ILC2s are inhibited by E-cadherin, and its downregulation recent studies showed that skin-resident.56

**NEUROIMMUNOLOGIC MECHANISMS**

A subset of sensory neurons that express histamine H1 receptor and histamine H4 receptor is activated by histamine, which can cause itch as well as allergic inflammation.57 H1 antihistamines have been widely used for the treatment of itch due to urticaria, but its effects are limited in the treatment of chronic itch in patients with AD. Recently, much interest has focused on the role of histamine-independent itch signaling pathways in which TSLP and type 2 cytokines, such as IL-4, IL-13, and IL-31, stimulate neurons expressing transient receptor potential cation channel subfamily A member 1 and afferent neurons via its receptors and Janus kinase (JAK) family, respectively.19 Of note, IL-31 induces sensory nerve elongation and branching,
which supports its role that involves sensitivity to minimal stimuli and sustained itch in patients with AD. In addition, the activation of STAT3 in the astrocytes of the spinal dorsal horn has been reported to be involved in chronic pruritus via the generation of lipocalin-2.

**EPIDERMAL DYSFUNCTION**

IL-4, IL-13, IL-31, IL-33, and high-mobility group box 1 downregulate the production of epidermal barrier proteins, including FLG, keratins, loricrin, involucrin, and cell adhesion molecules. A damaged epidermal barrier not only leads to the development of AD but also heightens sensitization to allergens and contributes to the risk of Food allergy (FA) and airway hyperreactivity. Impairment of skin barrier function at birth and at 2 months, as evaluated by transepidermal water loss (TEWL), can precede clinical AD by 12 months of age. Moreover, increased TEWL in the early newborn period is associated with a higher incidence of FA at 2 years of age, which supports the concept of transcutaneous allergen sensitization. Defects in epidermal barrier proteins, such as FLG, transglutaminases, keratins, and intercellular proteins, facilitate dysregulated immune responses to external antigens and drive skin and systemic inflammatory responses (Table 1). FLG is highly downregulated in both lesional and nonlesional skin of patients with AD.

Recently, McAleer et al. demonstrated that FLG breakdown products in the first year of life are lowest in the cheek compared with the elbow and the nasal tip, and the slowest to achieve maturity levels, which supports the importance of FLG on the pathogenesis of infantile AD. In that study, FLG processing enzymes such as bleomycin hydrolase and calpain-1 were also increased at cheek skin by 1 month of age. This may explain the predilection for AD at the cheeks initially in early childhood. Epidermal FLG levels are also reduced by environmental factors, including low humidity, sunburns, diesel exhaust particles, and skin irritants. In addition, loricrin and involucrin are downregulated by overexpression of Th2 cytokines through a STAT6-dependent mechanism in AD skin. Corneodesmosin (CDSN) and tight junctions play a central role by supporting the adhesion between corneocytes and the integrity of the skin barrier as an intercellular protein. A recent study showed that CDSN was downregulated by IL-4, IL-13, IL-22, IL-25, and IL-31 in human keratinocytes, and the penetration of vaccinia virus was enhanced in a CDSN-deficient skin model. In addition, claudin 1–deficient mice were reported to die...
within 1 day of birth with wrinkled skin appearance and severe dehydration, which provides good evidence for the essential role of claudin for the skin barrier function.\textsuperscript{72}

AMPs, including cathelicidin (LL-37) and human \( \beta \)-defensins, are produced by keratinocytes and play a pivotal role for host defense as well as control of host physiologic functions, such as inflammation and wound healing.\textsuperscript{73} AMP expressions are inhibited by Th2 cytokines, which are highly produced in AD skin.\textsuperscript{74} The decreased expression of AMPs is associated with a higher predisposition to \textit{Staphylococcus aureus} colonization, which can aggravate AD.\textsuperscript{75} It has been reported that human \( \beta \)-defensins and LL-37 are chemotactants for T lymphocytes, monocytes, dendritic cells, and neutrophils, and can induce cytokine production by monocytes and epithelial cells.\textsuperscript{76,77} These immunomodulatory properties of AMPs have important roles for host defense against infections through activation of immune cells as well as their direct antimicrobial activity.

**LIPIDS**

Lipids, such as ceramides, long-chain FFAs, and cholesterol, constitute the lipid matrix that is organized in lamellar bodies and located between corneocytes.\textsuperscript{78} During epidermal differentiation, precursor lipids are stored in lamellar bodies within the upper cell layers of the epidermis and extruded into the extracellular domain.\textsuperscript{79} Subsequent enzymatic processing produces the major lipid classes, which are necessary to maintain the integrity of the epidermal barrier. Altered lipid composition is observed in lesional and nonlesional AD skin.\textsuperscript{20} In particular, long-chain EO ceramides are essential because they are covalently bound to cornified-envelope proteins and cover the surface of each corneocyte.\textsuperscript{79} Th2 cytokines reduce levels of long-chain FFAs and EO ceramides with a STAT6-dependent manner.\textsuperscript{17,18,20} The levels of long-chain ceramides were decreased in patients with AD and who were colonized with \textit{S. aureus} when compared with those who were not colonized. TEWL was negatively correlated with levels of these ceramides.\textsuperscript{80}

**MICROBIOME**

AD skin has decreased bacterial diversity associated with increased \textit{Staphylococcus}, \textit{Corynebacterium}, and with reduced \textit{Streptococcus}, \textit{Propionibacterium}, \textit{Acinetobacter}, \textit{Corynebacterium}, and \textit{Propionibacterium} during AD flares.\textsuperscript{81,82} Greater bacterial diversity with increased abundance of \textit{Staphylococcus epidermidis} and \textit{Streptococcus}, \textit{Corynebacterium}, and \textit{Propionibacterium} species was observed after AD treatment and reduced eczema.\textsuperscript{82} Species-level investigation of AD has shown a higher predominance of \textit{S. aureus} in patients with more-severe disease and an abundance of \textit{S. epidermidis} in patients with less-severe disease.\textsuperscript{83} \textit{S. aureus} colonizes AD skin and has pivotal roles in the development and exacerbation of AD.\textsuperscript{84} \textit{S. aureus} can induce T-cell–independent B-cell expansion; upregulate proinflammatory cytokines, such as TSLP, IL-4, IL-12, and IL-22; and stimulate mast cell degranulation, which results in Th2 skewing and skin inflammation.\textsuperscript{85–88}

A recent study demonstrated that epidermal thickening and expansion of cutaneous Th2 and Th17 cells were induced when mice were exposed to \textit{S. aureus} isolates from patients with AD.\textsuperscript{83} Of note, methicillin-
resistant \textit{S. aureus} colonization on AD skin is associated with lower microbial diversity and a more profound reduction in the composition of commensal bacteria, such as \textit{Streptococcus} and \textit{Propionibacterium}, than methicillin-sensitive \textit{S. aureus} colonization.\textsuperscript{89} It is presumed that the differences and shifts in skin microbiome according to AD status are associated with the production of bacteriocins and AMPs from commensal bacteria.\textsuperscript{90,91} In addition, a recent study showed a positive correlation between the abundance of propionibacteria and corynebacteria on epidermis and long-chain unsaturated FFAs, such as FA20:1, FA20:2, FA22:1, and FA24:1.\textsuperscript{92} These findings highlight the importance of the balance between \textit{S. aureus} and commensal bacteria.

Patients with AD have significantly lower numbers of intestinal commensal \textit{Bifidobacterium} and higher numbers of \textit{Staphylococcus} than healthy control subjects.\textsuperscript{93} Overgrowth of pathogenic bacteria, such as \textit{Escherichia coli} and \textit{Clostridium difficile}, is postulated as being associated with a decrease in beneficial bacteria, reduced induction of regulatory T (Treg) cells, loss of immune tolerance, and increased intestinal permeability.\textsuperscript{94,95} These observations support the hypothesis that specific microbial composition in the gut prevents Th2-shifted immunity and stimulated regulatory immunity, producing regulatory dendritic cells and Treg cells.\textsuperscript{96,97} However, further studies are necessary to elucidate how dysbiosis affects epidermal barrier function and the development of AD.

**CLINICAL APPLICATION**

Frequent application of appropriate moisturizers, such as physiologic lipid mixtures and ceramide-dominant lipid, is known to help reduce TEWL, enhance skin hydration, decrease bacterial colonization, and improve skin barrier function, which leads to decreased need for topical corticosteroid.\textsuperscript{1,98,99} Petrolatum application has been reported to upregulate AMPs; induce key barrier differentiation markers, \textit{e.g.}, FLG; and reduces T-cell infiltration in AD skin.\textsuperscript{98} Of note, regular application of emollients has been reported to reduce the risk of AD development as a primary prevention strategy in infants at high risk.\textsuperscript{100,101} In addition, a recent study demonstrated that topical application of a liver X receptor agonist (VTP-38543) improved epidermal differentiation and lipids in patients with mild-to-moderate AD.\textsuperscript{102}

Topical calcineurin inhibitors, such as tacrolimus and pimecrolimus, inhibit calcineurin-dependent T-cell activation, which leads to downregulation of proinflammatory cytokines.\textsuperscript{99} Systemic immunosuppressants, including cyclosporine, methotrexate, and azathioprine, are used in patients with severe and difficult-to-treat symptoms.\textsuperscript{99} However, these drugs have limitations and adverse reactions. Therefore, various biologics to target polarized immune pathways have been newly developed for patients with moderate-to-severe AD. Although omalizumab did not show beneficial effects to treat AD,\textsuperscript{103,104} dupilumab, a humanized monoclonal antibody (mAb) to block IL-4 and IL-13, has been approved by the Food and Drug Administration.\textsuperscript{105,106} Clinical efficacy of dupilumab occurred without significant safety concerns in adult patients with AD.\textsuperscript{105,106} Clinical trials are also underway with dupilumab in pediatric populations (NCT02407756, NCT02612454, NCT03054428, NCT03346434, NCT03345914). Because the upregulation of Th17 and Th22 cytokines have been identified in patients with AD, the blockade of these pathways is being investigated by using secukinumab and a human monoclonal antibody against interleukin-22 (ILV-094; NCT02594098, NCT01941537). Moreover, Guttman-Yassky \textit{et al.}\textsuperscript{107} reported that an anti-IL-22 mAb (fezakinumab) showed clinical improvement in patients with severe AD.\textsuperscript{107}

A recent study also showed clear trends of therapeutic effects of ustekinumab, which is an IL-12/IL-23p40 antagonist, to suppress Th1, Th17, and Th22 immune activation in adults with moderate-to-severe AD.\textsuperscript{108} However, there was no significant difference between treatment and placebo groups in that study.\textsuperscript{108} Another Japanese study also did not demonstrate meaningful efficacy of ustekinumab on AD,\textsuperscript{109} although it is known to be effective for psoriasis.\textsuperscript{110} Nemolizumab (anti–IL-31R mAb), lebrikizumab (anti–IL–13 mAb), and tralokinumab (anti–IL–13 mAb) revealed promising results.\textsuperscript{106} Other biologic agents, such as Bristol-Myers Squibb-981164 (anti–IL–31 mAb), Tezepelumab (anti–TSLP mAb), and MK-8226 (anti–TSLP receptor mAb), are studied and may offer a range of new therapeutic options of AD. In addition, topical tofacitinib (JAK1/JAK 3 inhibitor) and oral baricitinib (JAK1/JAK2 inhibitor) were reported to have reduced skin inflammation and pruritus in patients with AD.\textsuperscript{111,112}

Although topical and systemic antibiotics have been used to eradicate bacteria from AD skin, long-term use has limitations due to the induction of resistant microorganisms and the negative impact on host commensal bacteria. Recent studies reported that a bleach bath is effective for the restoration of skin microbiome and the treatment of AD.\textsuperscript{113,114} However, a recent meta-analysis did not show its additional benefits compared with water bath alone.\textsuperscript{115} Interestingly, Nakatsuji \textit{et al.}\textsuperscript{116} found targeted autologous skin microbiome transplantation of \textit{S. hominis} and \textit{S. epidermidis} decreased \textit{S. aureus} from AD skin. Another recent study showed that the topical transplantation with \textit{Roseomonas mucosa} improved AD severity and reduced \textit{Staphylococcus aureus} colonization.\textsuperscript{117}

Recent studies demonstrated that appropriate probiotics are beneficial in the prevention and treatment of AD through the modulation of host immune re-
However, there have still been controversies regarding these clinical effects of probiotics in patients with AD, which might be due to a difference in the strains of probiotics and the characteristics of the host. It is noteworthy that the response to probiotics is greater in patients with an immunologically active state characterized by high total immunoglobulin E levels and increased expression of transforming growth factor β and Treg cells. Analysis of these emerging data indicated that identification of adequate AD phenotypes for the specific therapeutic option could be a key to achieve a good clinical outcome (Fig. 2).

CONCLUSION

Multiple factors, including epidermal gene mutations, skin barrier dysfunction, immune dysregulation, neuroinflammation, altered lipid composition, and microbial imbalance, can contribute to the development of AD. Various strategies have been used to restore skin barrier function and control skin inflammation in patients with AD. To overcome limitations of topical anti-inflammatory drugs and systemic immunosuppressants, substantial effort has been committed to the development of new therapeutic options, including biologics and microbiome transplantation. In addition, moisturizers and probiotics may prevent the development of AD in infants at high risk. Further advances in our understanding of AD pathophysiology will allow us to achieve a precision medicine approach to the prevention and the treatment of AD.

ACKNOWLEDGMENTS

We thank Samsung Medical Information and Media Services, Samsung Medical Center for the preparation of figures for this article.

REFERENCES

9. Egawa G, and Kabashima K. Multifactorial skin barrier deficiency and atopic dermatitis: Essential topics to prevent...


