Clinical reviews in allergy and immunology

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Deciphering the complexities of atopic dermatitis: Shifting paradigms in treatment approaches

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2. To review the role of cytokines in patients with AD.
3. To discuss new approaches in the treatment of AD.

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Atopic dermatitis (AD) is the most common chronic inflammatory skin disease. It often precedes the development of food allergy and asthma. Recent insights into AD reveal abnormalities in terminal differentiation of the epidermal epithelium leading to a defective stratum corneum, which allows enhanced allergen penetration and systemic IgE sensitization. Atopic skin is also predisposed to colonization or infection by pathogenic microbes, most notably *Staphylococcus aureus* and herpes simplex virus. Causes of this abnormal skin barrier are complex and driven by a combination of genetic, environmental, and immunologic factors. These factors likely account for the heterogeneity of AD onset and the severity and natural history of this skin disease. Recent studies suggest prevention of AD can be achieved through early interventions to protect the skin barrier. Onset of lesional AD requires effective control of local and systemic immune activation for optimal management. Early intervention might improve long-term outcomes for AD and reduce the systemic allergen sensitization that leads to associated allergic diseases in the gastrointestinal and respiratory tract. (J Allergy Clin Immunol 2014;134:769-79.)

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Atopic dermatitis (AD) is the most common chronic inflammatory skin disease.[1,2] Recent studies reveal strong associations between mental health disorders and AD, suggesting the need to effectively manage this disease for patient's general well-being and their family's quality of life.[3] AD is often associated with food allergy and asthma.[4,5] The abnormal skin barrier in patients with AD might allow epicutaneous absorption of environmental allergens through the skin and promote systemic allergen sensitization, which predisposes to the development of food allergy and asthma. In this month's issue of the *Journal*, researchers report

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Abbreviations used
AD: Atopic dermatitis
ADEH+: Atopic dermatitis with a history of eczema herpeticum
ADEH−: Atopic dermatitis without a history of eczema herpeticum
AMP: Antimicrobial peptide
DC: Dendritic cell
FLG: Filaggrin
TLR: Toll-like receptor
TMEM79: Transmembrane protein 79

that AD increases the effect of environmental peanut exposure in children with AD.[6-8] Because there are currently no cures for food allergy and asthma, the development of effective treatments for AD might be an important strategy for prevention of the atopic march. Therefore elucidation of the underlying mechanisms of AD provides a critical opportunity for early intervention.

AD is a complex disease with a genetic predisposition strongly influenced by innate and adaptive immune responses, as well as environmental factors, including allergen exposure, irritants, microbes, diet, stress, and air quality.[9-13] Although it is commonly referred to as a single disease,[14] recent studies suggest that the time has come to distinguish various AD phenotypes and endotypes[15,16] in much the same manner in which attempts have been made to categorize asthma and rhinosinusitis into different subtypes based on a constellation of the onset, biomarkers, immune polarization, gene variants, and natural history of the disease.[17-19] Identification of immune pathway polarity will be of particular importance as biologic agents become more readily available to target specific immune pathways, such as the T_H2 and T_H22 pathways, as well as various inflammatory cytokines and mediators.[20,21]

This month's issue of the *Journal* focuses particularly on the importance of both genetic and acquired causes of epithelial skin barrier dysfunction in driving the natural history of AD.[22-24] Two original articles this month report that early emollient use to protect the skin barrier might prevent AD.[25,26] Although dermatologists and allergists often debate the relative importance of genetic defects in the skin barrier giving rise to a leaky skin epithelial barrier that allows penetration of allergens and microbes into the skin of patients with AD (ie, the so-called "outside-in hypothesis" as opposed to the "inside-out hypothesis," a polarized immune response giving rise to a defective skin barrier), this argument is moot in patients with established AD because both processes are equally important (Fig 1). The majority of patients with AD constitute admixtures of genetic defects in the skin barrier and immune responses strongly influenced by environmental factors. This review will highlight recent insights into the crosstalk between the skin barrier and immune dysfunction leading to AD. Effective prevention and treatment of AD requires a multipronged approach involving the maintenance of skin barrier integrity, control of skin inflammation, nutrition, and identification and management of allergenic and microbial triggers.[27]

COMPLEX CAUSES OF EPITHELIAL SKIN BARRIER DYSFUNCTION IN PATIENTS WITH AD

Multifunctional role of filaggrin

The robust association of loss-of-function mutations in the skin barrier gene encoding filaggrin *(FLG)* with risk of AD has focused attention on the important role of epithelial barrier dysfunction in

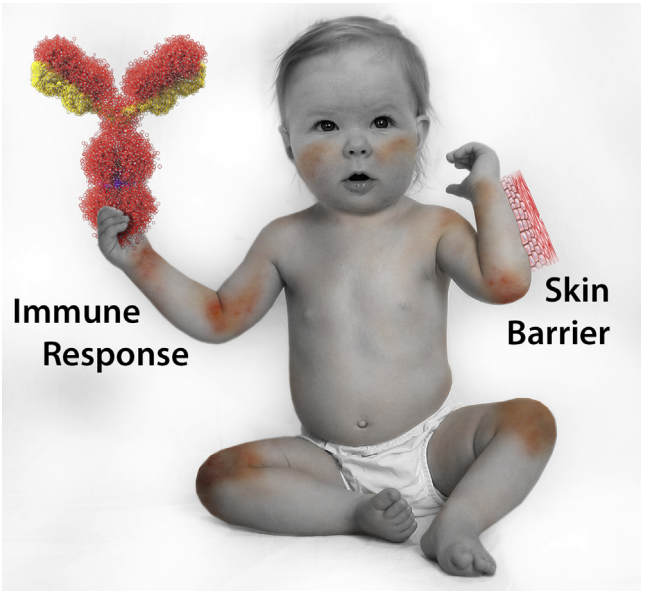

FIG 1. Is it clinically relevant whether skin barrier dysfunction or an immune response occurred first? Once AD is established, the physician needs to address both aspects of AD pathophysiology. However, prevention of AD might require identification of patients with primary defects in barrier versus immune dysfunction. Figure courtesy of Boyd Jacobson, National Jewish Health, Denver, Colorado.

TABLE I. Comparison of clinical and biophysical features of patients with AD with (AD_{FLG}) and without ($AD_{NON\text{-}FLG}$) filaggrin mutations*

	Clinical features	Biophysical features
AD_{FLG}	Palmar hyperlinearity	Severe decrease in NMF
	More persistent	pH
	↑ Allergic sensitization	IL-1β
	↑ Risk of asthma	Type 1 interferon–mediated stress response
	↑ Severity of AD	
	↑ Eczema herpeticum	
$AD_{NON\text{-}FLG}$	No palmar hyperlinearity	Mild decrease in NMF
	Less persistent	pH lower compared with patients with AD_{FLG}
	Less allergic sensitization	IL-1β low compared with patients with AD_{FLG}
	Lower risk of asthma	Dysregulation of lipid metabolic processes

NMF, Natural moisturizing factor.
*Modified with permission from McAleer and Irvine.[28]

patients with this skin disease.[28,29] Patients with filaggrin mutations have been found to have dry skin and early-onset AD that is more persistent and often associated with asthma, food allergy, and microbial infection.[30-32] Recent studies suggest that stratification of patients with versus without filaggrin mutations identifies patients with different mechanistic pathways of inflammation (Table I).[28] Patients with filaggrin loss-of-function mutations are associated with enhanced expression of IL-1 cytokines in their stratum corneum and type 1 interferon–mediated stress response.[33,34] Children with AD with normal filaggrin genes have been reported to have dysregulation of lipid metabolic processes.[34] Filaggrin-dependent secretion of sphingomyelinase has also been found to protect against staphylococcal

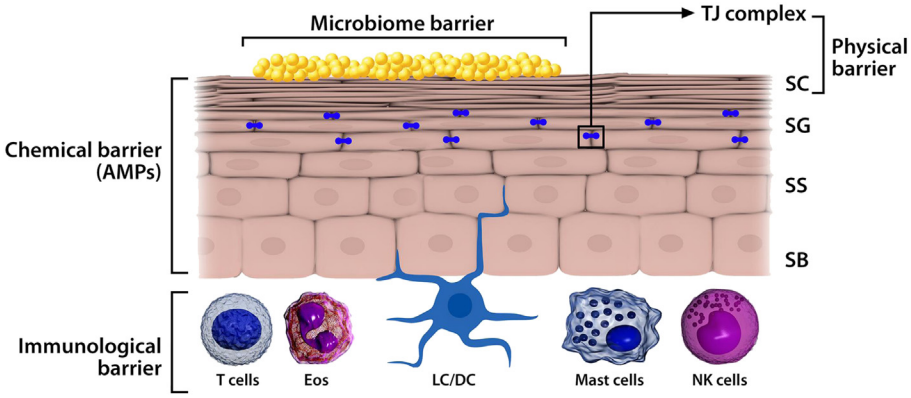

FIG 2. The skin as a multitiered barrier. The stratum corneum *(SC)* is the first physical barrier protecting the skin from the environment. Gene mutations (eg, filaggrin-null mutations) or cytokines (eg, IL-4, IL-13, IL-25, and IL-33) downregulating epidermal proteins, including filaggrin, leads to allergen or microbial penetration through this barrier. Tight junctions *(TJs)* found at the level of the stratum granulosum *(SG)* provide an additional barrier. Disruption of both physical barriers enables the uptake of allergens, irritants, and microbes by Langerhans cells *(LCs)*/DCs. Keratinocytes produce AMPs as a chemical barrier in response to pathogen colonization/infection. The skin surface is colonized by a diverse array of microorganisms (microbiome barrier) that dysregulate local immune responses and inhibit pathologic microbes. There is also infiltration of a number of cells into the AD skin lesion, including T cells, eosinophils *(Eos)*, DCs, natural killer *(NK)* cells, and mast cells/basophils. Collectively, these cells constitute the cutaneous immunologic barrier. Pattern recognition receptors regulate the function of all of these barriers (physical, chemical, microbiome, and immunologic). *SB*, Stratum basale; *SS*, stratum spinosum. This figure is modified from Kuo et al.[51]

α-toxin–induced keratinocyte death.[35] This strongly suggests that patients with filaggrin mutations have a distinct endotype of AD with different mechanistic outcomes (Table I), which could be used to identify one subset of AD, particularly for the development of new therapies targeting skin barrier function.

Clinical expression of AD is dependent on gene-environment and gene-gene interactions. Gene-environment interactions are best illustrated in filaggrin-deficient mice, in which exposure of the skin to allergens or microbes predictably leads to the development of eczema.[36] Three recent articles in the *Journal* demonstrate that environmental peanut might drive sensitization to peanut allergy in patients with AD, particularly those with filaggrin mutations, which is a clinically relevant example of the importance that environmental exposures in house dust might contribute to allergen sensitization in patients with AD.[6-8]

The importance of gene-gene interactions is illustrated in flaky tail mice, an animal model of AD with spontaneous eczema under pathogen-free conditions. These mice carry a double mutation involving the matted *(ma)* gene, giving them a matted hair phenotype, as well as a deletion in *Flg*. It was originally thought that the filaggrin deficiency in flaky tail mice explained the propensity of these mice to have AD. Surprisingly, the derivation of genetically engineered filaggrin-deficient mice that were free of the *ma* gene mutation were found to display impaired barrier function but to lack the propensity for spontaneous eczema.[37,38] The matted phenotype in flaky tail mice was found to be due to a loss-of-function mutation in the transmembrane protein 79 *(Tmem79)* gene. Unexpectedly, the *Tmem79* mutation, rather than the deletion in *FLG*, was found to be associated with the development of dermatitis in mice. Interestingly, *Tmem79* encodes lamellar granules that are required for processing of filaggrin, lipids, proteases, and antimicrobial peptides (AMPs).[22] Saunders et al[37] have also found that a single nucleotide polymorphism in the human *TMEM79* gene confers a significant risk for AD in human subjects, even when controlling for the effect of *FLG* mutations, suggesting both genes are involved in AD and the need for gene-gene interactions.

Depending on the population, *FLG* mutations are found in up to 40% of patients with severe AD, but less than 20% of these patients with severe disease are homozygous or compound heterozygous for *FLG* mutations.[39] Furthermore, only a minority of European American and Asian patients and none of the African American patients with AD have *FLG* mutations.[28,29,40,41] Reductions in filaggrin expression are also pronounced in the skin of patients with AD who have no detectable *FLG*-null mutations but are most profound when combined with *FLG* mutations.[42] Thus there are multiple causes for low *FLG* expression in the skin. The most common reason is likely to be immune activation.[42-44] Intragenic copy number variation within the filaggrin gene also contributes to the risk of AD with a dose-dependent effect.[45] The expression of *FLG* gene expression can also be reduced by means of epigenetic modification.[46]

Skin barrier dysfunction: Beyond filaggrin

Aside from *FLG*, AD has been associated with variants in other genes that encode a cluster of proteins in the epidermal differentiation complex located on chromosome 1q21.[47] These include filaggrin 2,[48] hornerin,[49] and the cornified envelope precursor SPRR3.[50] However, it is noteworthy that unlike *FLG*, the biologic function of these epidermal differentiation complex gene variants as it relates to AD is not well understood. However, a substantial amount of information indicates that loss-of-function mutations in serine protease inhibitors (eg, SPINK5) augment protease-activated pathways that enhance T_H2 responses, supporting the argument that epidermal barrier dysfunction can induce allergic skin diseases.[23] This complexity in epidermal gene variants is further modified by variants in genes that control innate and adaptive immune responses, as reviewed by Barnes[9] and Kuo et al.[51]

The normal skin can be viewed as containing a series of interrelated barriers the function of which is retention of moisture and repelling penetration of the skin by allergens and microbial invasion. Once the stratum corneum is breached, such as through a

deficiency of structural proteins (eg, filaggrin, involucrin, and loricrin), lipids (eg, ceramides), or both, other barrier structures are engaged (Fig 2). These include tight junction proteins, such as the claudins, which are found on opposing membranes of stratum granulosum keratinocytes directly below the stratum corneum and thereby form a second physical barrier in the epidermis.[51] Gene profiling in the epidermis of patients with AD has revealed downregulation of claudin protein and function in these patients. Once these 2 physical barriers (filaggrin and tight junctions) are breached, a rapid innate immune response must be initiated to prevent further microbial invasion. Keratinocytes and antigen-presenting cells in the skin express innate pattern recognition receptors, such as Toll-like receptors (TLRs). Stimulation of TLRs by microbes or tissue injury leads to the release of AMPs and enhanced strength of tight junctions to limit penetration of allergens and microbes.[51] Patients with AD have reduced TLR function.

Loss of skin barrier function and increased severity of AD predisposes to microbial colonization and chronic skin inflammation. This is due to increased expression of tissue receptors for *Staphylococcus aureus*, which leads to colonization of *S aureus* in atopic skin.[52,53] Keratinocytes from the skin of patients with AD have also been found to be deficient in their ability to produce the AMPs that are needed to control *S aureus* and viral replication.[54,55] Interestingly, commensal bacteria also produce AMPs capable of controlling *S aureus* growth.[56] *S aureus* produces high levels of serine proteases that can degrade the skin barrier.[57] Therefore an overabundance of *S aureus* in patients with poorly controlled AD can reduce barrier function through multiple mechanistic pathways.

Immune-mediated barrier dysfunction

Although there are strong arguments for the "outside-in" hypothesis suggesting that AD is fundamentally a disease of fixed (genetic) epidermal barrier defects,[22,23] there are equally compelling arguments that some forms of AD are primarily driven by polarized immune pathways that downregulate keratinocyte terminal differentiation, thereby creating a secondary skin barrier defect. The arguments against a primary role of the barrier defect in triggering keratinocyte hyperplasia and secondary immune activation include the following.

1. The *FLG* mutation is absent in most patients with AD.[28,29,58]
2. The majority of children with AD outgrow their disease, even in the presence of an *FLG* mutation.[59]
3. Unlike ichthyosis vulgaris, in which the entire skin is affected at birth, in the same genetic background patients with AD with *FLG* mutations have both lesional and nonlesional skin and the disease develops at some later time point and does not start at birth.
4. Both lesional and nonlesional AD skin exhibit a broad range of differentiation abnormalities beyond filaggrin (eg, loricrin, involucrin, corneodesmosin, and claudins), suggesting reactive epidermal differentiation/cornification alterations.[60,61]
5. Treatment of keratinocytes with IL-4, IL-13, IL-22, IL-25, and IL-31 directly downregulates filaggrin expression and increases kallikrein function, which can directly cause barrier dysfunction.[21,23,42-44,62,63] IL-22 directly induces keratinocyte hyperplasia and downregulation of filaggrin expression.[64,65]
6. Mice that are genetically engineered to overexpress T_H2 cytokines in their skin spontaneously have AD and *in vivo* skin barrier defects.[66-69]
7. Filaggrin expression is restored by using anti-inflammatory regimens with either topical calcineurin inhibitors or topical corticosteroids.[70]
8. The strongest argument is the resolution of clinical AD disease activity in patients with moderate-to-severe disease with broad-based immunosuppressive therapies, such as cyclosporine or narrow-band UV phototherapy,[71,72] and immune-targeted therapeutics (dupilumab), which is coupled with resolution of the abnormal epidermal responses.[20]

It is noteworthy that AD skin lesions are always associated with underlying immune activation.[61,73,74] In patients with chronic AD, several underlying features are invariably present: (1) increased skin infiltration by T cells (approximately 10-fold increase over background T-cell levels in normal skin); (2) increased skin infiltration by myeloid ($CD11c^+$) dendritic cells (DCs; also approximately 10-fold increase over normal skin levels), with most DCs having an "inflammatory phenotype" ($BDCA1^-/CD11c^+$)[75]; (3) increased production of cytokines and chemokines by activated T cells and DCs within skin lesions, as measured by means of quantitative mRNA measures for individual molecules and immunohistochemical detection of associated protein products in skin lesions; and (4) reactive epidermal hyperplasia or "regenerative maturation," showing an unusual hyperplasia response in which mRNAs and proteins of epidermal cornification are highly suppressed in the associated epidermis.[21,76-78]

Immune pathways driving AD skin lesions

In patients with AD with increased IgE levels, nonlesional AD is associated with a selective expansion of T_H2 cells in a dermal perivascular distribution.[73] The acute initiation of AD skin lesions is associated with T_H2, T_H22, and also T_H17 cytokine activation (Fig 3).[21] In parallel, there are epidermal (S100) responses marked by an extremely high increase in the expression of the proinflammatory epidermal differentiation complex cluster–encoded S100A genes (S100A7-9), which are known to be regulated by IL-22 and IL-17 cytokines.[65,79] In patients with chronic AD, intensification of T_H2 and T_H22 activation occurs with the appearance of a significant T_H1 component but not a complete switch to T_H1.[21] Although T_H2 cytokine effects remain during chronic AD, the increase in IFN-γ levels contributes to the inflammatory response and causes keratinocyte apoptosis.[80]

In the last decade, T_H2 and T_H22 cytokines have been reported to modulate the epidermal barrier, including suppression of keratinocyte differentiation, hyperplasia, keratinocyte apoptosis, and AMP production.[42-44,54,81-84] The cytokine effects include (1) suppression of terminal differentiation genes, such as filaggrin *(FLG)*, loricrin, and involucrin by T_H2 cytokines (IL-4, IL-13, and IL-31) and T_H22/IL-22 cytokines; (2) inhibition of AMP production by T_H2 cytokines (IL-4 and IL-13); (3) upregulation of S100As by IL-17 and IL-22[21,83-85]; and (4) induction of epidermal hyperplasia by the T_H22 IL-22 cytokine.[78]

AD disease activity (quantified by using the SCORAD score) has also been shown to positively correlate with lesional and nonlesional skin expression of T_H2 and T_H22 mediators (ie, IL-13 and IL-22) and negatively with expression of terminal differentiation markers.[71,81-84] Although immune activation is significantly higher in lesional than nonlesional skin, impressive reductions in

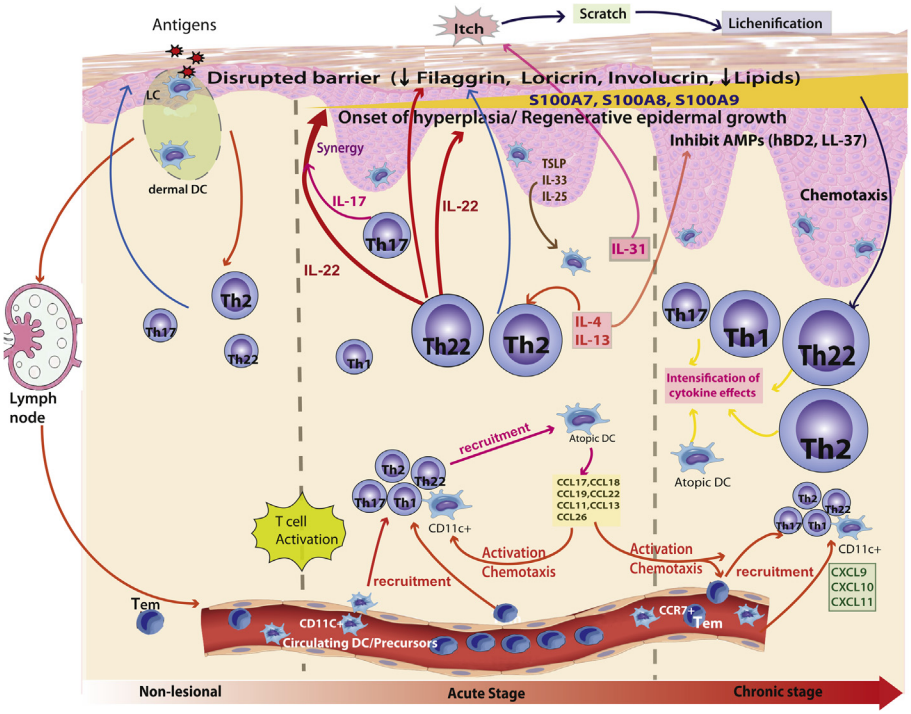

FIG 3. Immunologic pathways involved in different phases of AD. Nonlesional AD skin lesions contain immune infiltrates that produce cytokines, such as IL-4 and IL-13, which contribute to a defective epidermal barrier. Barrier defects lead to penetration by epicutaneous allergens that encounter Langerhans cells in the epidermis and dermal DCs in the dermis to activate T_H2 and T_H22 cells involved in acute disease onset. Smaller increases in T_H1 and T_H17 immune axes are also found in acute lesions. A progressive activation of T_H2 and T_H22, as well as T_H1, pathways is characteristic of patients with chronic AD. IL-22 induces epidermal hyperplasia and, synergistically with the T_H17 cytokine IL-17, drives an abrupt increase in a subset of terminal differentiation genes, specifically S100A7, S100A8, and S100A9 proteins. The increases in levels of these barrier proteins contrast with the uniformly disrupted epidermal differentiation gene products (eg, filaggrin, loricrin, and corneodesmosin) throughout nonlesional, acute, and chronic AD skin. The T_H2 and T_H22 cytokines contribute to inhibition of the terminal differentiation proteins. IL-31 is thought to contribute to the itch in patients with acute AD. *TSLP*, Thymic stromal lymphopoietin. Updated with permission from Gittler et al.[21]

expression of a broad array of epidermal differentiation genes (ie, loricrin, periplakin, and involucrin in addition to filaggrin) characterize both lesional and nonlesional AD skin. Because T_H2 (IL-4/IL-13) and T_H22 (IL-22) cytokines have been shown to inhibit expression of terminal differentiation products in keratinocytes, increased circulating levels of these cytokines might cause this global suppression of barrier proteins, as well as increases in levels of S100As detected at the onset of acute lesions.[21]

IMPLICATIONS OF AD PATHOBIOLOGY FOR GENERAL MANAGEMENT APPROACHES

Patients with established AD have a combination of skin barrier dysfunction and skin inflammation driving their skin disease. Therefore keys to the successful management of AD should include skin hydration and skin barrier repair, topical anti-inflammatory medications (topical corticosteroids or calcineurin inhibitors), control of infection, and elimination of factors (including allergens, irritants, and emotional triggers) that might exacerbate the scratch-itch cycle. Treatment should use a stepwise approach that is dependent on the severity of skin disease. The reader is referred to several recent excellent reviews on the management of AD.[27,85-87]

In managing patients with chronic AD, it is important to recognize that gene profiling and immunohistologic studies reveal subclinical inflammation and downregulation of terminal epithelial differentiation with reduced skin barrier protein levels and increased transepidermal water loss, even in nonlesional AD skin.[47,61,74,87,88] Thus it is important to maintain skin barrier therapy in the form of emollient therapy, even during periods of remission. In patients with AD who are prone to frequent relapses, the subclinical inflammation can be managed with intermittent (2 times per week) corticosteroids or alternate-day topical calcineurin inhibitors as maintenance therapy.[89] For acute AD exacerbations, medium- and high-potency corticosteroids can be used for short periods of time to control the disease. Oral or systemic corticosteroids should be avoided because of rebound flares when patients are being weaned from oral corticosteroids.

In patients with AD who are refractory or do not clinically respond to conventional treatment approaches, a number of alternative strategies can be used, including cyclosporine, methotrexate, azathioprine, IL-6 blockade, dust mite immunotherapy (when indicated), wet wrap therapy, and UV light.[90-94] Recent studies with broad-based targeting therapeutics[71,72] have used disease-related cellular and molecular biomarkers to (1) show that the chronic AD phenotype can be reversed to a nonlesional state, as has been shown for patients with psoriasis treated with effective therapeutics, and (2) map inflammatory disease-related pathways. The narrow-band UVB phototherapy and cyclosporine trials showed elimination of the pathologic epidermal hyperplasia

(suprabasal K16 expression immunohistochemically and increased K16 and Ki67 mRNA expression) after 12 weeks of treatment. The improvement in disease activity as identified by using the SCORAD score and epidermal pathology were highly linked to clearance of excess T-cell and DC infiltrates, as well as decreased expression of inflammatory markers.[71,72,95,96]

PREVENTION OF AD BY EARLY INTERVENTION

Because current treatment approaches are not curative, there is considerable interest in studying approaches to prevent AD.[97] The use of probiotic therapy or bacterial lysates early in the course of illness to prevent AD remains an area of active investigation,[98-100] but results have been inconsistent. This might be due to lack of standardization of the bacterial preparations or lack of biomarkers to identify which AD phenotype would benefit from this approach.

The potential contribution of vitamin D deficiency to allergic inflammation, corticosteroid insensitivity, and downregulation of innate immune responses has also been an active area of study.[101-106] Reproducible well-controlled studies of oral vitamin D supplementation are lacking, and when they have been done, they have yielded confusing results. The greatest benefits are likely in populations who have extremely low vitamin D levels, such as persons living in upper latitudes during the winter or darkly pigmented persons. In the current issue of the *Journal*, Camargo et al[13] present results from a randomized, placebo-controlled trial demonstrating that winter-related AD can be improved with vitamin D oral supplementation.

The importance of AD skin barrier dysfunction in driving allergen sensitization is highlighted by 3 articles in the current issue suggesting that severe AD drives sensitization with environmental peanut.[6-8] In 2 of these articles, filaggrin mutations predicted increased association of AD with peanut allergy. These articles suggest the possibility that controlling environmental peanut levels in the household might reduce peanut allergen sensitization. Alternatively, studies are needed to determine whether effective control of AD with barrier therapy and anti-inflammatory treatment to reduce AD skin severity will reduce absorption of environmental allergens and decrease onset of food allergies or respiratory allergy.

Considering the important role that skin barrier dysfunction plays in the initiation of AD, the current issue of the *Journal* contains 2 different international investigations assessing early intervention with skin emollient therapy to prevent AD and allergic sensitization during infancy.[25,26] Simpson et al[25] performed a randomized controlled trial of 124 neonates at high risk for AD. Parents in the intervention arm were instructed to apply full-body emollient therapy at least once per day starting within 3 weeks of birth. Parents in the control arm were asked to use no emollients. The primary outcome was the cumulative incidence of AD at 6 months. Their results demonstrated a statistically significant protective effect with the use of daily emollient on the cumulative incidence of AD, with a relative risk reduction of 50%.

Horimukai et al[26] performed a randomized controlled trial with early moisturizer intervention conducted in 116 neonatal participants at high familial risk for AD. The primary outcome was the cumulative incidence of AD as of week 32, as evaluated by a dermatologist. The intervention with the moisturizer significantly decreased (by approximately 40%) the risk of AD compared with that seen in the control subjects ($P = .002$) as of week 32. The 2 groups showed similar rates of allergic sensitization. However, the rate of allergic sensitization of infants with AD was significantly higher than that seen in the rest of the infants.

These 2 studies suggest that early intervention with emollient therapy from birth represents a safe and effective approach for AD prevention. If confirmed to be effective in future studies, emollient therapy from birth would be a simple and low-cost intervention that could reduce the global burden of allergic diseases. Whether this form of intervention can prevent the atopic march is unresolved and might require combination with intermittent anti-inflammatory therapy and environmental control.

TABLE II. Clinical phenotypes of AD

- Onset in infancy, outgrown in childhood
- Onset in infancy, persistent severe eczema
- Adolescent-adult onset, mild-to-moderate eczema
- Adolescent-adult onset, persistent severe eczema
- Increased IgE levels with food or aeroallergen sensitization (extrinsic)
- Non-IgE mediated (intrinsic)
- AD with *S aureus* infection/colonization
- AD with history of disseminated viral infections (eg, eczema herpeticum)

CLINICAL PHENOTYPES OF AD

AD is primarily defined by clinical criteria.[107] However, there is increasing recognition that AD is a complex syndrome with multiple causes and mechanistic pathways that clinically can be distinguished by age of onset, severity of illness, racial modifiers, response to therapy, and triggers (including infections, allergens, stress, and irritant threshold). Table II lists some of the major clinical phenotypes of AD.[15,16] These phenotypes often have overlapping features but contain dominant characteristics that distinguish them from each other (Fig 4).[108] The majority of infants who present with mild AD will outgrow their skin disease in later childhood. However, a group of difficult-to-manage patients exist who have early-onset eczema with severe lifelong AD. Adult-onset AD has also been increasingly reported, although it is unclear whether these might be patients who had eczema during infancy and then went into a prolonged remission only to have relapse of eczema later in life because recall history can be unreliable. Up to 50% but certainly not all patients with AD have associated asthma, allergic rhinitis, or food allergy. Identification of genetic markers and biomarkers of patients likely to undergo the atopic march would allow early intervention for prevention of mucosal allergy, including food allergy and asthma.

Approximately 80% of patients with AD have increased serum IgE levels and/or immediate skin test reactivity to allergens, but 20% of patients with AD have no IgE to food or inhalant allergens. However, it is possible that such intrinsic or nonatopic patients might have IgE or autoreactive T cells to autoallergens or microbial antigens, which are not routinely measured.[109-111] Other AD subsets exist, including those who are prone to skin infection, such as *S aureus* skin infections or eczema herpeticum.[112-116] Although up to 90% of patients with AD might have problems with *S aureus* skin colonization, actual overt skin infections requiring systemic antibiotic treatment affect less than 50%. Less than 3% of patients with AD are predisposed to eczema herpeticum. These different phenotypes likely arise from a complex combination of mutations and epigenetic effects on genes controlling protein expression in the skin barrier and the innate and adaptive immune response controlled by environmental influences.[9,117]

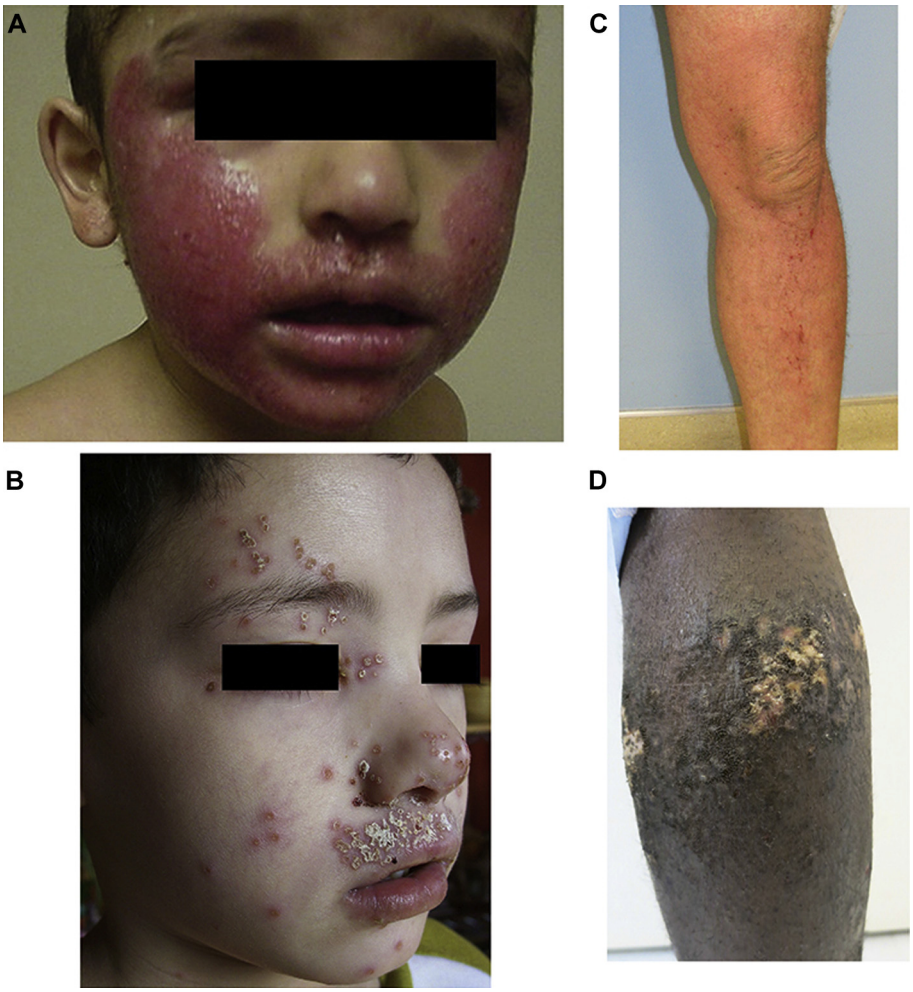

FIG 4. Clinical phenotypes in patients with AD: eczema herpeticum **(A)**, *S aureus*–colonized AD **(B)**, mild AD **(C)**, and severe AD **(D)**. Fig 4, *A* and *B*, are from Boguniewicz and Leung.[108] Fig 4, *C* and *D*, were contributed by Dr Emma Guttman-Yassky at the Icahn School of Medicine at Mt Sinai, New York.

TABLE III. Summary of cytokine effects on the epidermis in patients with AD

- Induce epidermal hyperplasia (IL-22)
- Induce spongiosis (T_H2 cytokines IL-4/IL-13 and TNF)
- Inhibit keratinocyte terminal differentiation (IL-4, IL-13, IL-31, IL-25/T_H2, IL-22/T_H22, and TNF) with potential for feedback hyperplasia
- Inhibit synthesis of AMPs (T_H2 cytokines IL-4, IL-13, and IL-33)
- Inhibit lipid synthesis (T_H2 cytokines IL-4/IL-13, IL-31, and TNF)
- Increase expression of S100A7, S100A8, and S100A9 (IL-22 plus IL-17)
- Induce TSLP production in KCs (IL-4/IL-13 and TNF)
- Promote itch (IL-31 and TSLP)
- Promote antiviral responses (IFN-γ, IFN-α, and IL-29)

KCs, Keratinocytes; *TSLP*, thymic stromal lymphopoietin.

DEFINING ENDOTYPES IN PATIENTS WITH AD

The importance of eventually defining endotypes in patients with AD is that these new subtypes can be used in clinical study design and drug development to target therapies to patients most likely to benefit from a mechanism-based treatment. In the future, AD might be stratified by genotype and biomarkers reflecting immune polarization to complement their clinical phenotype. As noted in Table I, filaggrin genotyping defines AD subsets with different mechanistic pathways. Importantly, the severity of AD is related to filaggrin expression with a dose-dependent effect.[45]

Patients with AD with homozygous filaggrin-null mutations or compound heterozygotes compared with patients with normal filaggrin gene expression have early onset of skin disease and more persistent severe eczema. They often have palmar hyperlinearity, greater risk of allergen sensitization, and a history of food allergy and asthma. In addition, they have increased pH values in their stratum corneum, which might predispose them to *S aureus* colonization. Patients with heterozygous *FLG* mutations have an intermediate phenotype and can outgrow their AD in adolescence, whereas homozygotes or compound heterozygotes can have lifelong disease.[59] These patients might also serve as a target for filaggrin therapy.[118]

Given the complex genetic milieu of AD, the development of biomarkers is important to assess the final immune-polarized pathways that might exist in various AD subsets. The best biomarkers for AD currently define patients who are T_H2 polarized versus those who are not. In the future, a combination of epidermal proteomics, genomics, gene transcriptomes, and blood biomarkers in combination with the clinical phenotype will offer more precision in defining endotypes of AD.[119-121]

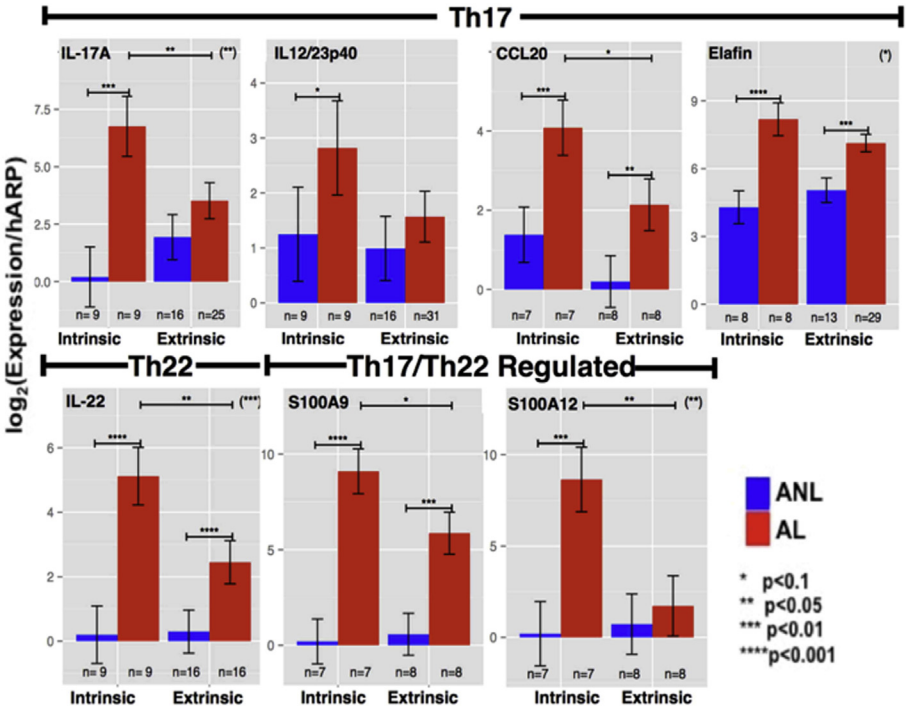

FIG 5. Measures of mRNA levels (normalized to hARP mRNA) for specific T_H17/IL-23 and T_H22 cytokines and inflammatory products in nonlesional *(ANL)* and lesional *(AL)* AD skin from patients with intrinsic versus extrinsic disease. From Suarez-Farinas et al.[124]

Approximately 80% of patients with AD have increased serum IgE levels, often with increased eosinophilia and serum levels of the T_H2 chemokine thymus and activation-regulated chemokine. Additional markers are needed to better monitor patients with so-called intrinsic AD. However, it is noteworthy that studies of patients with so-called intrinsic AD who lacked IgE to conventional inhalant and food allergens had detectable serum IgE to autoantigens in the skin and microbial antigens from bacterial and fungi that colonize the skin.[110,111] Therefore a wider range of IgE screens to various exogenous and endogenous antigens is warranted to determine potential triggers of AD because it might have an important effect on pathways triggering allergic skin inflammation.

Elucidation of the mechanisms underlying *S aureus* and disseminated viral infections in patients with AD is an active area of investigation. These patients are generally very atopic, with increased serum IgE levels and eosinophilia. This might reflect high-level T_H2 cytokine pathway activation, which is known to reduce skin barrier function, enhance *S aureus* skin colonization, reduce AMP production, and impair innate immune responses. However, because eczema herpeticum is extremely rare and herpes simplex virus exposure is very common, it is likely that additional immunologic and genetic factors contribute to atopic dermatitis with a history of eczema herpeticum (ADEH+). To identify novel gene signatures, in the current issue of the *Journal*, Bin et al[122] used an RNA-sequencing approach to evaluate global transcriptional changes in PBMCs from patients with ADEH+ and those with atopic dermatitis without a history of eczema herpeticum. They found that PBMCs from patients with ADEH+ stimulated with herpes simplex virus 1 were deficient in their antiviral immune response involving interferon regulatory factor 3 and 7 innate immune pathways. This likely contributes to the reduced interferon response in ADEH+ that predisposes to increased susceptibility to disseminated viral infection. Interestingly, dedicator of cytokinesis 8 deficiency, which presents with eczema and recurrent herpetic skin infections, has recently been found to respond well to treatment with IFN-α 2b.[123]

Disease severity appears to be related to the magnitude and polarity of immune activation, as well as effects of immune cytokines on epidermal responses (Table III).[21,61,95,96,124] Severe AD might thus be associated with systemic immune activation, including significant pathology in nonlesional skin,[61] explaining the frequent need for systemic immune suppressants and the inadequacy of treating only lesional skin with topical agents. In contrast, because of minimal systemic involvement, it might be most appropriate to treat mild disease with topical agents directed only to lesional sites.

Immune studies suggest that different AD phenotypes are associated with distinct patterns of activation (or suppression) of polar immune axes and corresponding tissue responses. There are distinct differences in cytokine production in patients with intrinsic versus extrinsic AD.[124] In patients with intrinsic AD, who have normal IgE levels, there is significantly increased expression of IL-17, IL-23, IL-22, and their respective keratinocyte-induced products (ie, S100As, elafin/PI3, and CCL20), suggesting a subset with potential for greater responsiveness to suppression of the IL-17/IL-23/IL-22 axes (Fig 5).[124] Despite high IgE levels in patients with extrinsic AD, similar expression levels of T_H2-related products were observed in both patients with extrinsic and those with intrinsic AD, suggesting that these phenotypes might have similar responses to IL-4 receptor antagonism. Indeed, Beck et al[20] showed that responses to dupilumab treatment were similar in patients with intrinsic and those with extrinsic AD.

TABLE IV. Phenotypes of severity and treatment response

- Preclinical: Use skin barrier cream for prevention of AD
- Chronic AD in remission: Use barrier cream in combination with maintenance topical corticosteroid or calcineurin inhibitors to prevent relapse
- Relapse of mild-to-moderate AD: Use topical corticosteroids or calcineurin inhibitors for control of inflammation, identify and avoid triggers (irritants, allergens, and infection), immunotherapy for allergen-driven AD
- Persistent moderate-to-severe AD not controlled with topical corticosteroids or calcineurin inhibitors: Wet wrap therapy, allergen immunotherapy, and nonspecific immunosuppressives (cyclosporine, methotrexate, narrow-band UV phototherapy, and mycophenolate)
- Future targeted therapies for moderate-to-severe AD (anti–IL-4 receptor α, anti–IL-22, anti–IL-23/IL-17, or other biologic agents that interrupt polarized immune pathways)

Strong support for the changing pathogenic and therapeutic AD paradigm comes from a recent report on the use of IL-4 receptor α antagonist/dupilumab conducted in patients with moderate-to-severe AD.[20] This study showed major improvement (approximately 70%) in disease activity at the higher dose (compared with only approximately 20% in the placebo group) coupled with large improvement in the AD genomic phenotype and reversal of the epidermal hyperplasia (as quantified by larger reductions in K16 expression).[20] In fact, the reduction in hyperplasia was much higher in the 4-week trial than with 5 mg/kg cyclosporine given for 12 weeks to patients with similar disease activity.[95]

CONCLUDING COMMENTS: THE TRANSLATIONAL REVOLUTION IN AD

AD presents a large unmet need for more effective topical and systemic therapeutics. In addition to T_H2 antagonists (ie, anti–IL-4 receptor/dupilumab), the key role of thymic stromal lymphopoietin receptor signaling[125,126] and IL-22,[127] as studied in clinical trials with agents targeting thymic stromal lymphopoietin, T_H22, and T_H17/IL-23, will be of interest. Selection of immune-targeted therapeutics for patients with different degrees of disease severity or recognized AD phenotypes should not be done by serendipity but should be guided by defining the extent of activation of polar immune circuits in skin and blood (Table IV). For example, anti–IL-23/IL-17 might provide beneficial responses in patients with AD, particularly patients with intrinsic AD. The individual contributions of the T_H22, T_H17, and T_H2 immune pathways to the disease phenotype will be clarified through clinical trials coupled with mechanistic studies that are currently in progress.

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