Atopic Dermatitis, Pathophysiology: New Insights and Therapeutic Implications
Mark Boguniewicz, MD
Professor, Division of Allergy-Immunology
Department of Pediatrics
National Jewish Health and University of Colorado School of Medicine
Denver, Colorado USA

Objectives
At the end of this session, participants will be able to:
• Discuss new insights into the pathophysiology of atopic dermatitis including skin barrier and immune abnormalities
• Understand implications of skin barrier abnormalities for allergic diseases

Eczema Prevalence in the United States*


* data from 2003 National Survey of Children’s Health

Global variations in prevalence of eczema symptoms in children from ISAAC Phase Three


Atopic March and beyond: The prevalence of atopic triad in children with physician-confirmed atopic dermatitis

• In the largest cross-sectional study of a cohort of 2270 children with diagnosis of AD confirmed by physician and treated for a minimum of 6 wks with topical prescription product for their AD:
  – nearly 66% had symptoms of asthma or allergic rhinitis and 38% had both associated with their AD
  – nearly 80% reported some additional allergic illness (asthma, allergic rhinitis, seasonal allergy, food allergy, animal allergy, or drug allergy) by the third year of life
• Results differ from previous studies in that prevalence rates of asthma and allergic rhinitis are higher and the onset is earlier i.e., if an additional illness is to be noted, it will be by the age of 3 years

Complex immune dysregulation

IL-22-producing “T22” T cells account for upregulated IL-22 in atopic dermatitis despite reduced IL-17-producing TH17 T cells

A milestone in the genetics of complex allergic disorders…

Common loss-of-function variants of the epidermal barrier protein filaggrin are a major predisposing factor for AD

• Filaggrin (filament-aggregating protein) is a key protein that facilitates terminal differentiation of the epidermis and formation of the skin barrier
• FLG gene located on chromosome 1q21 in the EDC
• 2 loss-of-function genetic variants (R510X and 2282del4) in FLG shown to be very strong predisposing factors for AD
• These variants also show highly significant association with asthma occurring in the context of AD
• This data suggests a key role for impaired skin barrier function in development of atopic disease

Filaggrin in atopic dermatitis

Genes contributing to skin barrier breakdown


Tight junctions and claudin-1: A new barrier defect in atopic dermatitis?

- Reduced expression of TJ protein claudin-1 in AD pts validated at mRNA & protein levels
- Claudin-1 expression inversely correlated with Th2 biomarkers
- Functional relevance associated with impairment of bioelectric barrier function in AD
- CLDN1 SNPs associated with AD


FILAGGRIN

- FLG null alleles occur in up to 50% of patients with moderate-severe AD suggesting a fundamental role for barrier homeostasis in this disease
- Patients with FLG mutations more likely to have early-onset, severe, persistent AD
- FLG mutations are the strongest and most widely replicated genetic risk factors for AD identified to date

Variation in filaggrin mutations among ethnic groups and other populations

- FLG null alleles associated strongly with eczema; eczema associated with these mutations presents in early life and is more persistent (hazard ratio for eczema resolution for those with FLG mutations to FLG wild type, 0.67; 95% CI, 0.58-0.77; P = 5 x 10^-8)


Skin barrier function and allergic risk

- FLG mutations conferred a population asthma risk of 1.80 (95% CI, 1.34-2.41; P = .00019); asthma risk was especially high in the context of eczema (odds ratio, 3.16; 95% CI, 2.25-4.43; P = 1.4 x 10^-11)
- Strong associations were identified with sensitization to grass, house dust mite, and cat dander and sensitization to multiple allergens (odds ratio, 2.12; 95% CI, 1.03-4.37; P = 5.42 x 10^-2)
- FLG mutations confer risk of a particular trajectory for eczema, with increased duration of disease and greater risk of asthma and multiple allergic sensitizations
- FLG alleles help define the risk profile of children with eczema and help define the “eczema plus early wheeze” and “eczema plus asthma” phenotypes

Hudson TJ. Nature Genetics 2006;38:399

The burden of disease associated with filaggrin mutations: a population based, longitudinal birth cohort study

- To determine the natural history and burden of atopic disease conferred by the 2 most common FLG mutations in a large, population-based birth cohort study
- Analyzed the effect of the most common null alleles (R501X and 2282del4) on several atopic phenotypes in a cohort of approximately 2000 English children born in 1990-1991
- FLG null alleles associated strongly with eczema; eczema associated with these mutations presents in early life and is more persistent (hazard ratio for eczema resolution for those with FLG mutations to FLG wild type, 0.67; 95% CI, 0.58-0.77; P = 5 x 10^-8)

**Loss-of-function variants in the filaggrin gene are a significant risk factor for peanut allergy**

- **Case-control study of 71 English, Dutch, and Irish oral food challenge–positive pts with peanut allergy and 1000 non peanut-sensitized English population controls**
- **Replication tested in 390 white Canadian pts with peanut allergy (defined by food challenge or clinical history and skin prick test wheal to peanut ≥ 8 mm and/or peanut-specific IgE ≥ 15 kU/L) and 891 white Canadian population controls**
- **Most prevalent filaggrin loss-of-function mutations assayed in each population: R501X and 2282del4 in Europeans, and R501X, 2282del4, R2447X, and S3247X in Canadians**


**Meta-analysis of filaggrin polymorphisms in eczema and asthma: robust risk factors in atopic disease**

- **FLG mutations significantly increase risk of**
  - AD, odds ratio 3.12; 95% CI 2.57-3.79
  - Asthma, odds ratio 3.29; 95% CI 2.84-3.82
- **Associated with more severe AD**


**Filaggrin gene defects and risk of developing allergic sensitisation and allergic disorders: systematic review and meta-analysis**

- **24 studies**
- **Odds of developing allergic sensitization was 1.91 (95% CI 1.44 to 2.54) in family studies and 1.57 (1.20 to 2.07) in case-control studies**

Filaggrin is a robust biomarker for allergic conditions

*Data supports an Atopic March*

Filaggrin haploinsufficiency and increased risk of several complex traits


Recent insights into atopic dermatitis and implications for management of infectious complications

Increased susceptibility to infections or colonization with microbial organisms: *Staphylococcus aureus, Herpes simplex*


*S. aureus* adherence to normal healthy skin and uninvolved AD skin


*S. aureus, toxins & AD*

- Most AD pts colonized by toxin-secreting *S. aureus*
- ~25% isolates from AD pts MRSA (higher ~50% in Southern states)
- Both MSSA & MRSA produce alpha-toxin critical for enhanced survival on skin
- ~10% of *S. aureus* isolates from AD pts shown to produce 2 superantigenic toxins (SEB & TSST) (co-production not seen in isolates from >5000 pts with TSS)
- Use of topical steroids and TCIs may select for *S. aureus* strains that adapt to their “new” skin surface niche by increasing virulence capabilities
- AD pts make serve as an important reservoir for evolving *S. aureus* strains

A deficiency in antimicrobial peptide* (H-β-defensin-2) expression may allow *S. aureus* to colonize and infect skin of AD patients.

*keratinocytes are the major source of AMPs in the skin*


**Effects of IL-4 and IL-13 on TNF-α-induced H-β-defensin-2 expression in HaCat cells**

![Graph showing effects of IL-4 and IL-13 on TNF-α-induced H-β-defensin-2 expression in HaCat cells.](chart)

Medium

<table>
<thead>
<tr>
<th>Condition</th>
<th>Expression (pg/μg rRNA)</th>
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<tr>
<td>TNF-α</td>
<td>1.2 ± 0.2</td>
</tr>
<tr>
<td>TNF-α + IL-4</td>
<td>9.3 ± 0.4</td>
</tr>
<tr>
<td>TNF-α + IL-13</td>
<td>11.5 ± 0.5</td>
</tr>
<tr>
<td>TNF-α + IL-4 + IL-13</td>
<td>12.0 ± 0.5</td>
</tr>
</tbody>
</table>

p < 0.05


**IL-4 and IL-13 inhibit cathelicidin expression through STAT-6 and can be reversed by neutralizing antibodies**


**Thymic stromal lymphopoietin:** Master switch for allergic inflammation

![Diagram showing Thymic stromal lymphopoietin](chart)

**Phenotype of atopic dermatitis subjects with a history of eczema herpeticum**

- AD patients with more polarized Th2-type disease with allergies and asthma and increased biomarkers including serum IgE, TSLP and cutaneous T cell-attracting chemokine more likely to have severe skin disease complicated by eczema herpeticum, *S. aureus* or molluscum infections


**FLG mutations and Th2 polarization**

*Genetic variants in thymic stromal lymphopoietin are associated with AD & EH*

- A critical link between barrier defect in AD patients with FLG mutations and Th2 polarization could be explained in part by enhanced allergen penetration through the damaged epidermis accompanied by increased production of TSLP* by keratinocytes leading to a Th2-type milieu.
- Significant associations for TSLP and IL7R tagging SNPs and AD and ADEH in European Americans with replication of associations between TSLP and IL7R SNPs in an independent African American sample.


**Filaggrin mutations that confer risk of atopic dermatitis confer greater risk for eczema herpeticum**

- 2 common loss-of-function mutations plus 9 FLG single nucleotide polymorphisms genotyped in 278 European American patients with AD, of whom 112 had ADEH, and 157 nonatopic controls; replication performed on 339 African American subjects.
- Significant associations observed for both the R501X and 2282del4 mutations and AD among European American subjects (P = 1.46 x 10(-5), 3.87 x 10(-5), respectively), but the frequency of the R501X mutation was 3 times higher (25% vs 9%) for ADEH than for AD without EH (OR, 3.4; 1.7-6.8; P = .0002).
- Associations with ADEH stronger with the combined null mutations (OR, 10.1; 4.7-22.1; P = 1.99 x 10(-11)).


**NIAID Atopic Dermatitis Research Network**

- Research area 2: Susceptibility to staphylococcal colonization and infections, including MRSA.
  - Clinical Mechanistic Studies:
    - Identification of genetic determinants for bacterial dissemination in ethnically diverse patients with AD.
    - Identification of epidermal barrier defects that predispose AD subjects to colonization with *S. aureus*.
    - Exploratory studies on PBMC immunoprofiles in AD subjects with and without bacterial colonization.
  - Clinical Trial:
    - AD, innate immunity and vitamin D.
  - Animal Mechanistic Study:
    - *S. aureus*, innate immunity and vitamin D in mouse models.

**ADRN S. aureus related research**

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Research area 3: Susceptibility to colonization and/or infection with other commensal organisms.

- Clinical Mechanistic Study of the skin microbiome in AD patients to understand the susceptibility to colonization and/or infection with other commensal organisms.
- Registry & Repository of Biological Specimens.
- *S. aureus* repository.

*Bojarski JW, et al. In Pediatric Allergy 2010: 986*
**TH2 cytokines downregulate filaggrin gene expression**


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**Different effects of pimecrolimus and betamethasone on the skin barrier in patients with atopic dermatitis**

- 15 AD pts randomized to pimecrolimus (P) to one upper limb and betamethasone (S) to other 2x/day for 3 wks
- Stratum corneum (SC) hydration and TEWL improved and dye penetration reduced with both treatments
- EM evaluation of barrier structure showed prevalently ordered SC lipid layers and regular lamellar body extrusion in P-treated skin but inconsistent extracellular lipid bilayers and only partially filled lamellar bodies with B treatment
- Both drugs normalized epidermal differentiation and reduced epidermal hyperproliferation
  - Decreased filaggrin restored with both treatments
- B was superior in reducing clinical symptoms and epidermal proliferation; however, it led to epidermal thinning


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**Psoriasis patients treated with a TNF-alpha antagonist* showed increased protein expression of filaggrin...**


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**Nonlesional atopic dermatitis skin is characterized by broad terminal differentiation defects and variable immune abnormalities**


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**Reversal of atopic dermatitis with narrow-band UVB phototherapy and biomarkers for therapeutic response**