Cutaneous microbiome effects of fluticasone propionate cream and adjunctive bleach baths in childhood atopic dermatitis

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Background: Patients with atopic dermatitis (AD) are prone to skin infections, with microbes such as *Staphylococcus aureus* suspected of contributing to pathogenesis. Bleach baths might improve AD by reducing skin microbial burden.

Objective: We sought to characterize the microbiota of lesional and nonlesional skin in young children with AD and control subjects and compare changes after treatment with a topical corticosteroid (TCS) alone or TCS + dilute bleach bath.

Methods: In a randomized, placebo-controlled, single-blinded clinical trial in 21 children with AD and 14 healthy children, lesional and nonlesional AD skin was examined at baseline and after 4-week treatment with TCS alone or TCS plus bleach bath. Microbial DNA was extracted for quantitative polymerase chain reaction of predominant genera and 16S rRNA sequencing.

Results: At baseline, densities of total bacteria and *Staphylococcus*, including *Staphylococcus aureus*, were significantly higher at the worst AD lesional site than nonlesional ($P = .001$) or control ($P < .001$) skin; bacterial communities on lesional and nonlesional AD skin significantly differed from each other ($P = .04$) and from control ($P < .001$). After TCS + bleach bath or TCS alone, bacterial compositions on lesional skin normalized ($P < .0001$), resembling nonlesional skin, with microbial diversity restored to control skin levels.

Limitations: The 4-week time period and/or the twice-weekly baths may not have been sufficient for additional impact on the cutaneous microbiome. More detailed sequencing may allow better characterization of the distinguishing taxa with bleach bath treatment.

Conclusions: Treatment with a TCS cream suffices to normalize the cutaneous microbiota on lesional AD; after treatment, bacterial communities on lesional skin resemble nonlesional skin but remain distinct from control. (J Am Acad Dermatol 2016;75:481-93.)

Key words: atopic dermatitis; bleach baths; cutaneous microbiome; quantitative polymerase chain reaction; 16S sequencing; *Staphylococcus aureus*; topical corticosteroids.

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Atopic dermatitis (AD) is a common, chronic inflammatory skin condition characterized by pruritic eczematous lesions in specific distribution patterns in infants and children.[1] Morbidity in patients with AD is often caused by cutaneous infections with particular bacteria, fungi, and viruses. *Staphylococcus aureus* is highly prevalent on AD skin, and culture-based studies have shown direct correlation between AD clinical severity and *Staphylococcus aureus* density.[2] Whether *Staphylococcus aureus* contributes to the pathophysiology or merely reflects the abnormal environment has never been conclusively established. Molecular techniques enable a more comprehensive characterization of the changing cutaneous microbiota in skin disease and its relationship to clinical features.[3,4]

CAPSULE SUMMARY

- Atopic dermatitis (AD) flares are associated with increases in *Staphylococcus aureus* density.
- Microbial communities on nonlesional AD skin are distinct; topical corticosteroids normalize the cutaneous microbiome on both nonlesional and lesional AD skin.
- The addition of twice weekly bleach baths to standard topical steroid treatment with fluticasone propionate cream is not required for clinical improvement or normalization of microbial communities on AD skin.

In this study, in the context of a clinical trial comparing topical corticosteroids (TCS) alone with TCS plus dilute bleach baths for AD treatment, we used high-throughput DNA sequencing and quantitative polymerase chain reaction (qPCR) to characterize the cutaneous microbiota of children with AD and control subjects. Comparison of lesional and nonlesional skin with that of controls at baseline and over the course of the 2 regimens enabled a more comprehensive view of microbiota characteristics and their changes with disease activity. Our results show that in addition to its clinical efficacy in treating AD, TCS treatment also normalizes cutaneous microbiota compositions in AD, even without added bleach bath.

METHODS

Patients and study design

Children of age 3 months to 5 years with moderate to severe AD (based on modified Hanifin and Rajka criteria as reported by Eichenfield et al[5]) were enrolled from the pediatric dermatology clinics at the Charles C. Harris Skin and Cancer Unit and New York University (NYU) Dermatologic Associates, both at NYU Langone Medical Center (NYULMC), and at Bellevue Hospital Center. Control patients were enrolled from the adjacent general pediatric clinics if they were in the correct age group, and did not have any skin disease. This study was approved by the NYULMC Institutional Review Board.

Exclusion criteria

Patients with concurrent chronic inflammatory skin disorders or who were currently using or had used systemic or topical antibiotics, corticosteroids, or calcineurin inhibitors for AD in the prior 2 weeks were excluded from the study, as were children with overt infection. See Fig 1 for study design.[6]

Randomization

Treatment randomization was done by 2 independent nonclinical staff members. One person made 12 sealed envelopes containing the word "bleach" and 12 identical sealed envelopes containing the word "water," which were then shuffled and numbered sequentially. The envelopes were opened in number order and plain white bottles were filled with the corresponding ingredient, bleach or water, and then labeled with the corresponding number.

Sampling

All patients with AD had 4 clinical sites swabbed including the worst affected site (ie, the lesion with the highest local Eczema Area and Severity Index [EASI] score), a nonlesional site, and 2 other representative lesional sites. Control patients were swabbed at the typical age-specific areas of eczema appearance. When possible, the nonlesional sample was taken from a site contralateral to the lesional site; however, when the presence of symmetric disease rendered this impossible, we selected an adjacent uninvolved area. Specimens were obtained using a sterile cotton-tipped applicator premoistened with 0.9% normal saline and 0.1% Tween 20 (Fisher Scientific, Waltham, MA), and swabbing the skin at the specified site with moderate pressure for 40 seconds, as exactly described.[7] After centrifugation, the fluid was used for DNA extraction, using the PowerSoil kit (MoBio, Carlsbad, CA) per the manufacturer's protocol.

Quantitative polymerase chain reaction

TaqMan qPCR (Applied Biosystems, Foster City, CA) was performed using specific primers and probes for the 16S rRNA sequences of *Staphylococcus aureus*, other *Staphylococcus* species, *Corynebacterium*, *Propionibacterium*, *Streptococcus*, and universal bacteria exactly as described.[8] *Staphylococcus aureus* was