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25 Keywords: food allergy, *Staphylococcus aureus*, eczema, peanut, egg, milk, IgE, skin, nasal,
26 environment

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28 Disclosure of potential conflict of interest: JMC-M, MJT, KMK, and RK declare that they have no
29 relevant conflicts of interest. MHK is a consultant for Janssen Vaccines.

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31 Food allergies often begin early in life. Although there is no cure for food allergies, recent
32 efforts to desensitize by immunotherapy have shown promise in lowering risk during accidental
33 exposures. In a retrospective study, early introduction of milk and peanut associates with
34 reduced incidence of allergy to milk ¹ and peanut ², respectively. In the prospective LEAP
35 (Learning Early about Allergy to Peanut trial) and LEAP-On studies ³, introduction of peanut to
36 4-11 month old children reduced risk of developing peanut allergy by age 5. However, 9% of
37 children were excluded at the selection phase because they were already sensitized to peanut
38 (defined by wheal size in skin prick test). Also, 7 of the 319 were excluded because they had an
39 oral response to peanut on initial challenge, and 9 of the 319 of the peanut consumer group
40 developed an oral response to peanut. Because peanut reactions can be life threatening and
41 reactions can occur during initial food challenge and oral immunotherapy protocols, it is critical
42 to identify risk factors for developing peanut allergic reactions.

43 A number of host extrinsic and intrinsic factors have been implicated in food allergy
44 sensitization. Host intrinsic factors associated with development of food allergy include skin
45 barrier function gene mutations and atopic eczema. Extrinsic factors predisposing to food
46 allergy include exposure to food and pro-inflammatory microbes and microbial components.
47 Skin colonization by *Staphylococcus aureus* (*S. aureus*) associates with eczema severity and
48 the presence and severity of atopic eczema are both risk factors for food allergy. In children with
49 atopic dermatitis, skin colonization with *S. aureus* and elevated specific IgE (sIgE) to peanut,
50 egg, and milk correlate with a greater than 95% positive predictive value of oral food challenge
51 reactions ⁴. Atopic dermatitis and allergies are also associated with fungal and house dust mite
52 exposure ⁵. House dust is a ubiquitous important environmental source of these extrinsic
53 factors including *S. aureus*, food allergens, house dust mite (HDM) and *Alternaria alternata* (*Alt*)
54 (Figure 1A).

55 Preclinical studies suggest that these environmental skin exposures can induce food
56 allergy and may prevent induction of oral tolerance to food allergens. In neonatal mice with skin

57 barrier gene mutations, food allergy develops after only four-40 minute skin co-exposures to
58 peanut or egg, *Alt* or house dust mite extract (HDM), and detergent to increase allergen
59 adsorption⁶. This allergen sensitization occurs in the absence any clinical evidence of atopic
60 eczema-like skin inflammation, which typically develops months later and in a manner
61 independent of allergen exposure. Similar to the human LEAP studies, development of food
62 allergy in mice can be blocked by pre-exposure to oral peanut before skin sensitization⁶.
63 However, not all neonatal mice are protected from developing food allergy. For example,
64 exposure of skin to *Alt* during the oral PNE pre-exposure blocked the oral peanut induced
65 tolerance⁶. This emphasizes how environmental factors can limit induction of tolerance during
66 consumption of food allergens. In addition, reports indicate mouse skin exposure to SEB with
67 food allergen (OVA or peanut) induces a Th2 phenotype and food allergy⁷. Another relevant
68 environmental exposure for the skin is detergent that decreases epithelial barrier function⁸.
69 Thus, impaired skin barrier function (induced by detergents and intrinsic genetic defects) in
70 combination with exposure of the skin to food allergens with meals and dust containing HDM,
71 *Alt*, *S. aureus* and food allergens likely synergize to promote development of food allergy
72 (Figure 1A). Various preclinical data support the idea that *S. aureus* colonization and other
73 environmental factors can predispose to food allergy and interfere with attempts to induce oral
74 tolerance to selected allergens.

75 In the current issue of the *Journal*, Olympia Tsilochristou et al. examined associations
76 between *S. aureus* skin/nasal colonization, food allergy and atopic eczema in LEAP and LEAP-
77 On study participants (Figure 1B). As expected, no difference in *S. aureus* colonization (skin or
78 nasal) was seen between LEAP peanut consumer and avoider subgroups. While there have
79 been studies in humans and murine models which have shown that antibiotic courses were
80 associated with development of food allergy, in the current study there were no associations of
81 prior oral or topical antibiotic use with *S. aureus* colonization. This suggests that prior antibiotic
82 use is not confounding the current associations with *S. aureus* colonization, skin barrier and

83 development of peanut allergy. Similar to other studies, concurrent *S. aureus* skin colonization
84 was associated with increasing atopic eczema severity. In addition, prior colonization with skin
85 *S. aureus* was associated with persistent or worsening of eczema severity at later time points.
86 Notably, prior skin colonization with *S. aureus* at 0 to 60 months of age was associated with
87 increased peanut, hen's egg white and cow's milk sIgE in a manner independent of total IgE,
88 eczema severity or severity of infection and across the entire cohort. In the entire cohort,
89 participants with prior skin or nasal *S. aureus* colonization had a 2.9 fold or 2.4 fold increased
90 odds, respectively, for peanut allergy at 60 months of age, regardless of eczema severity. For
91 peanut allergy, subgroup analysis of peanut consumers showed that there was a 7-fold
92 increased odds of peanut allergy with prior skin *S. aureus* colonization. Persistence of peanut
93 or egg allergy at 60 and 72 months of age was more likely in participants with skin or nose *S.*
94 *aureus* colonization prior to 0-60 months of age, regardless of eczema severity. Nasal *S. aureus*
95 association with food allergy was not as strong an association as skin *S. aureus*. Persistence
96 of egg allergy was associated with skin colonization of *S. aureus*, irrespective of peanut
97 ingestion. A potential clinical implication is that the protective effect of peanut ingestion on
98 peanut allergy was less marked in those with *S. aureus* colonization. For those patients that
99 failed induction of tolerance to peanut, skin exposures to *S. aureus* or other environmental
100 factors that are associated with *S. aureus* may have contributed to development of peanut
101 allergy. Thus, these LEAP and LEAP-On analyses demonstrate that early life cutaneous skin
102 exposure to *S. aureus* and early life eczema associate with subsequent food allergy
103 development, including peanut consuming children who developed food allergy during early
104 introduction of peanut.

105 A limitation to the study was that evidence for *S. aureus* was based on bacteriological
106 culture techniques rather than DNA-based testing. This approach may explain the lower
107 prevalence of cutaneous and nasal *S. aureus* colonization compared to prior studies⁹. In
108 addition, this detection method does not genotype the isolated strains and cannot detect

109 nonviable components of *S. aureus* like SEB and *S. aureus* antigens that act as immune
110 stimulants. A second limitation to consider is the small number (n=9) of study participants in the
111 peanut consumption group of LEAP and LEAP-On that developed peanut allergy. As noted by
112 the authors, prospective collection of larger numbers of subjects will be needed to further
113 investigate the relationship between *S. aureus* colonization and development of peanut allergy.
114 Also, as discussed by the authors, even though eczema was adjusted for and there were
115 associations of *S. aureus* with food allergy to peanut or egg, it does not exclude that other
116 environmental factors that associate with *S. aureus* can function in the development of food
117 allergy.

118 In summary, the current longitudinal prospective study by Tsilochristou et al¹⁰ provides
119 convincing evidence for an association between prior *S. aureus* colonization and food allergy to
120 egg or peanut, independent of atopic eczema severity (Figure 1B). The study suggest that *S.*
121 *aureus* colonization or factors associated with *S. aureus* colonization act as adjuvants that block
122 induction of oral tolerance and promote sensitization to allergens introduced via epicutaneous
123 exposure. Moreover, it suggests that *S. aureus* colonization may negatively impact clinical
124 approaches to induce oral tolerance to peanut and potentially, other allergens, in some patients.
125 It would be important to determine if attempts to induce oral tolerance to peanut, egg or other
126 allergens in *S. aureus* colonized subjects is augmented by eradication of *S. aureus* colonization.
127 Given the number of LEAP consumers who developed peanut allergy was small, larger studies
128 will be needed to determine whether there is a consistent association of food allergy with prior
129 *S. aureus* colonization. The current study has helped define new questions and consideration of
130 novel approaches (e.g. *S. aureus* skin decolonization) that may ultimately improve efforts to
131 induce oral tolerance.

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165 Figure Legend

166 Figure 1. **Skin Co-exposures in development of Food Allergy. A)** Preclinical models suggest
167 that a combination of skin exposures induce Th2 inflammation and food allergy. Skin is exposed
168 to food allergens, environmental allergens, and detergents. Many of these components are in
169 household dust. Household dust contains food allergens, HDM, *Alt*, and *S. aureus*. Also, *S.*
170 *aureus* is present on household surfaces. Skin exposure to food allergens also occur during
171 meals. Adsorption of allergens is promoted by detergents left on the skin from wet wipes and
172 cleansing products. Combinations of these components and genetic skin barrier mutations
173 readily induce food allergy. **B)** In LEAP and LEAP-On, skin *S. aureus* colonization levels
174 associate with eczema severity. Prior skin colonization with *S. aureus* associates with food
175 allergy and sIgE, but this is independent of eczema severity. There is an association of *S.*
176 *aureus* skin colonization with food allergy even after adjustment for eczema and total IgE. Thus,
177 there is an association of skin *S. aureus* with food allergy in children.

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