## Allergy in Children in Hand Versus Machine Dishwashing

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**BACKGROUND AND OBJECTIVE:** The hygiene hypothesis stipulates that microbial exposure during early life induces immunologic tolerance via immune stimulation, and hence reduces the risk of allergy development. Several common lifestyle factors and household practices, such as dishwashing methods, may increase microbial exposure. The aim of this study was to investigate if such lifestyle factors are associated with allergy prevalence.

**METHODS:** Questionnaire-based study of 1029 children aged 7 to 8 years from Kiruna, in the north of Sweden, and Mölndal, in the Gothenburg area on the southwest coast of Sweden. Questions on asthma, eczema, and rhinoconjunctivitis were taken from the International Study of Asthma and Allergies in Childhood questionnaire.

**RESULTS:** Hand dishwashing was associated with a reduced risk of allergic disease development (multivariate analysis, odds ratio 0.57; 95% confidence interval: 0.37–0.85). The risk was further reduced in a dose-response pattern if the children were also served fermented food and if the family bought food directly from farms.

**CONCLUSIONS**: In families who use hand dishwashing, allergic diseases in children are less common than in children from families who use machine dishwashing. We speculate that a less-efficient dishwashing method may induce tolerance via increased microbial exposure.

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WHAT'S KNOWN ON THIS SUBJECT: Microbial exposure during early life may prevent, or reduce, the risk of allergy development.

WHAT THIS STUDY ADDS: Allergic diseases are less common in children whose parents use hand dishwashing instead of machine dishwashing, and we hypothesize that this allergy-preventive effect is mediated via an increased microbial exposure.

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Asthma, eczema, and allergic diseases are common chronic conditions in children, and the rising prevalence during the later parts of the 20th century<sup>1</sup> led to intense research into possible causes. Initially, research focused on single causes, such as the increase in automobile traffic and air pollution,<sup>2</sup> damp buildings,<sup>3</sup> or allergen exposure.<sup>4</sup> However, knowledge accumulated from the large number of studies undertaken indicates that there is probably no single cause and no simple explanation. In different countries and settings, different risk factors and protective factors for asthma and allergy seem to interact, and the effect of this interaction on a specific individual depends on their genetic/ epigenetic susceptibility. We know, for example, that early day care attendance may protect against sensitization, but only in children without siblings.<sup>5</sup> Similarly, there is no additive allergy-protective effect from siblings in young adults who have serological markers of earlier infection with hepatitis A virus, Toxoplasma gondii, or Helicobacter *pylori*.<sup>6</sup> It has also been shown that an increased risk for sensitization in children from air pollution mainly occurs in genetically susceptible individuals.<sup>7</sup> This complicated interactive pattern sometimes makes general hypotheses difficult to interpret. Nevertheless, there are a few allergy-protective factors that seem to be active in most settings, even though their protective effect may differ depending on interactions with other environmental exposures. One of these unifying factors is immune stimulation by early microbial exposure. With early microbial exposure, there is both direct evidence of an allergyprotective effect<sup>8</sup> and indirect support for a dose-response effect.9 However, this protective effect may not be sufficient to maintain a low prevalence of allergy in all settings. There are many places in Africa and South America where allergy

prevalence is high despite an environment with many opportunities for microbial exposure.<sup>10</sup> This does not mean that early microbial exposure is not important in these countries, because we cannot tell what the prevalence would be if exposure to just that specific factor was reduced or delayed.

The hygiene hypothesis stipulates that microbial exposure during early life induces immunologic tolerance via immune stimulation, and hence reduces the risk of allergy development. In this epidemiologic study, we wanted to explore daily-life exposure patterns, presumed to affect microbial exposure, to test if these exposure patterns are associated with less allergy development in children. We postulated that there is an allergy-preventing effect associated with dishwashing practices and dietary habits; lifestyle factors that, if proven, could be used in allergy prevention in affluent countries.

#### **METHODS**

In September 2007, a questionnaire was sent to the parents/guardians of all children aged 7 to 8 years resident in Kiruna or Mölndal. Kiruna is a town in the north of Sweden; Mölndal is an integrated part of Gothenburg, a city on the southwest coast of Sweden. The Swedish version of the International Study of Asthma and Allergies in Childhood (ISAAC) questionnaire was used,<sup>11</sup> with permission from Professor Bengt Björkstén, to estimate the prevalence of asthma, rhinoconjunctivitis, and eczema.

In the current study, disease diagnoses were defined as follows:

- A diagnosis of eczema was based on the response to the ISAAC question "Has your child ever had eczema?"
- Asthma is difficult to separate from "episodic viral wheeze" in preschool-aged children; to increase the validity of the asthma

diagnosis (ie, to choose those with a high risk of persistent asthma), we included eczema in the definition of asthma.<sup>12–14</sup> A child was reported as having asthma if there was a positive response to the 2 ISAAC questions: "Has your child ever had asthma?" and "Has your child ever had eczema?"

- Allergic rhinoconjunctivitis (ARC) is often difficult to separate from infectious rhinitis in preschoolaged children due to the high frequency of common colds in this age group. The label ARC in the current study was therefore based on symptoms experienced in the last year; that is, if they gave a positive response to the ISAAC question "In the past 12 months, has your child had a problem with sneezing, or a runny, or a blocked nose when he/she DID NOT have a cold or the flu?" and at least 1 of the following: "Has your child ever had hay fever?" or "In the past twelve months, has this nose problem been accompanied by itchy-watery eyes?", or if symptoms occurred between April and August (the Swedish pollen season).
- Parental history of allergy corresponded to "a doctor's diagnosis" of asthma, ARC, eczema, or food allergy, as reported in the questionnaire.
- "Allergic disease" and "allergic diseases" are used in the text as an umbrella term for eczema, asthma, or ARC. When used as a variable name in the analyses, "total allergy" corresponds to any of eczema, asthma, or ARC based on the abovementioned disease definitions.

Five questions related to dietary or eating habits were tested. Translated versions of the questions in the Swedish questionnaire, with final steps used in the analysis, are given below.

• Dishwashing practices: Question: "How do you usually wash the dishes?" Responses: hand dishwashing; machine dishwashing.

- Fermented food: Question: "Does the child eat food that includes fermented vegetables (such as sauerkraut or fermented cucumber) or other fermented foodstuffs?" Responses: never/ almost never; at least once a month.
- Food from farm: Question: "Do you sometimes buy hens' eggs, meat, or unpasteurized milk directly from a farm?" Responses: yes; no.
- Home cooking: Question: "During the child's first year of life, how often was he/she given homecooked food?" Responses: never/ almost never; approximately half the time; most of the time/always.
- Breastfeeding: Question: "For how long was your child breastfed?" Response: duration in months.

"Traditional cooking" is used as an umbrella term in the text and analyses. The term includes any of dishwashing by hand, use of fermented food, and food from farm, as defined above.

The analysis included 5 major steps.

A principal component analysis (PCA) was used in the first step to explore the data.

In the second step, selected patterns were further tested in univariate analyses with  $\chi^2$  and Fisher's exact test. Differences are also expressed as odds ratios (ORs) with 95% confidence intervals (CIs).

The third step was used to identify possible confounding variables. In 3 orthogonal partial least squares (OPLSs) analyses, we analyzed possible associations between all independent variables and allergy, hand dishwashing, and traditional cooking, respectively. Variables that revealed a significant correlation with both total allergy and hand dishwashing, and total allergy and traditional cooking, respectively, were regarded as confounders.

In the fourth step, the confounders were entered into backward multiple logistic regression-models. In these 2 models, we also included parental history of allergy, pet keeping during infancy, and town code, because these variables were associated with total allergy. The complete list entered into the backward multiple logistic regression models were as follows: parental history of allergy, mother or father born in Sweden, mother's education, town code, pet keeping during infancy, day care attendance, crowding (square meters/number of persons in the home), mother smoking during pregnancy, and hand dishwashing (model 1) or traditional cooking (model 2), respectively.

In the fifth step, the variables from the last steps in model 1 and model 2 were used in hierarchical multiple logistic regression models. The hierarchical model analyzing the association between hand dishwashing and the different allergic diseases (model 3) adjusted for parental history of allergy, day care attendance, and pet keeping during infancy. The hierarchical model analyzing the association between traditional cooking and total allergy (model 4) adjusted for parental history of allergy, day care

| TABLE 1 | Study Population | Characteristics | (n = 1029) |
|---------|------------------|-----------------|------------|
|---------|------------------|-----------------|------------|

attendance, pet keeping during infancy, and father born in Sweden.

SIMCA software (version 13.0.3; Umetrics AB, Umeå, Sweden) was used for the PCA and OPLS analyses. Variables were centered and scaled to unit variance. SPSS software (version 22.0.0.0; IBM SPSS Statistics, IBM Corporation) was used for crosstabulations and multiple logistic regressions.

A written consent was obtained from the parents. The study was approved by the Human Research Ethics Committee of the Medical Faculty, University of Gothenburg, Sweden (Dnr 105-07).

#### **RESULTS**

Of the 1838 questionnaires distributed in Mölndal (n = 1354) and Kiruna (n = 484), 1029 (56%) were returned: 717 (53%) from Mölndal and 312 (64%) from Kiruna (Table 1). The population of children was well balanced with respect to gender. Most parents were born in Sweden. Educational level, as well as a parental history of allergy, was

| Characteristic   | No. of Respondents (%) |
|--|------------------------|
| Воу  | 483 (47)               |
| Resident in Kiruna                                     | 312 (30)               |
| Mother   |                        |
| Born in Sweden   | 899 (87)               |
| College/university degree                              | 510 (50)               |
| History of allergy                                     | 498 (48)               |
| Father   |                        |
| Born in Sweden   | 886 (86)               |
| College/university degree                              | 385 (37)               |
| History of allergy                                     | 399 (39)               |
| Child had siblings                                     | 952 (92)               |
| Smoking exposure                                       |                        |
| Mother smoked during pregnancy                         | 62 (6)                 |
| Smoking inside house during child's first year of life | 37 (4)                 |
| Pet exposure   |                        |
| Pets during pregnancy                                  | 294 (29)               |
| Pets during first year of life                         | 293 (28)               |
| Attended day care                                      | 940 (91)               |
| Current residence                                      |                        |
| Flat   | 240 (23)               |
| Detached, semidetached, or row house                   | 783 (76)               |
| Farm   | 6 (<1)                 |
| Dishwashing practices                                  |                        |
| Hand dishwashing                                       | 126 (12)               |
| Machine dishwashing                                    | 868 (84)               |

slightly higher in mothers than in fathers. Three of 4 families lived in a detached/semidetached house or a row house. Almost all children had siblings; most children had attended day care. Keeping of pets during early life was common, reported for almost one-third of the children, whereas few children were exposed to tobacco smoke.

The association between different exposure variables and allergy development was analyzed with PCA (Fig 1). One of the stronger associations was seen between dishwashing by hand and allergic disease (Tot\_all). The figure reveals a negative correlation between these 2 variables.

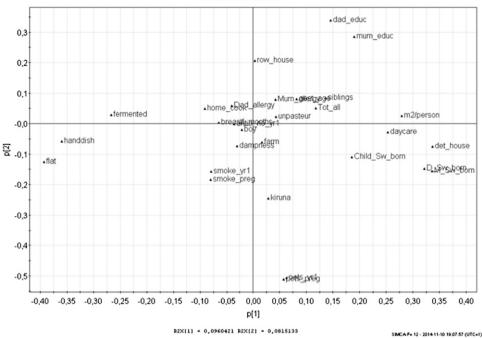
This negative correlation between hand dishwashing and specific allergic diseases, as well as possible associations between allergy and other dietary and eating habits were further tested in univariate analyses (Table 2). A history of eczema was

reported by 23% in children whose parents used hand dishwashing, whereas it was 38% in those who mainly used machine dishwashing (P = .001). The corresponding figures for asthma were 1.7% and 7.3%, respectively (P = .011). A similar pattern was seen for ARC (10.3% and 12.9%), but the difference was not statistically significant (P = .413). Associations between allergic diseases and 4 other lifestyle-related dietary habits are also presented in Table 2. Both eating fermented food and buying food directly from farms revealed a tendency toward protection against specific allergic diseases, but the difference was only statistically significant on the global level (ie, for total allergy).

Dishwashing by hand might, however, be associated with different lifestyle and socioeconomic factors that could act as confounders, explaining the lower prevalence of allergy seen in children whose parents use hand dishwashing. To further explore the association, we adjusted for covariates and confounding factors in multiple logistic regression-models (model 1 and model 3), as described in the Methods section. In Fig 2, the associations between hand dishwashing and specific allergic diseases are given, expressed as adjusted OR and 95% CIs. As shown in the figure, the associations between hand dishwashing and allergic diseases remain statistically significant for eczema and total allergy, whereas a borderline significance is seen for asthma.

To evaluate possible associations between allergic disease and cooking and dishwashing habits, we created the umbrella term traditional cooking (hand dishwashing, eating fermented food, or buying food directly from farms). Allergic disease, expressed as total allergy, was reported by 33% of those who used traditional cooking as compared with 46% of those who did





#### **FIGURE 1**

PCA loading plot, visualizing all included variables on 2 axes: those with the largest variation along the horizontal (first component) axis, and those with less variation along the vertical axis (second component). Approximately 18% (9.60% + 8.15% = 17.8%) of the total variability is described by the model. Variables located close together in the same quadrant are positively correlated. Variables located in diagonally opposed quadrants are negatively correlated. One of the stronger associations is seen between dishwashing by hand (handdish) and allergic disease (asthma, eczema, or ARC, labeled as Tot\_all in the figure). In the figure, handdish is located in the lower left quadrant and Tot\_all in the right upper quadrant, indicating a negative correlation between these 2 variables.

| Dietary Habit               | No. of Responses | Crude OR (95% CI) |                  |                     |                                       |
|-----------------------------|------------------|-------------------|------------------|---------------------|---------------------------------------|
|                             |                  | Eczema, $n = 368$ | Asthma, $n = 66$ | ARC, <i>n</i> = 130 | Total Allergy, <sup>d</sup> $n = 432$ |
| Fermented food <sup>a</sup> | 84               | 0.61 (0.37-1.01)  | 0.90 (0.35-2.31) | 0.52 (0.22-1.21)    | 0.53 (0.32-0.87)                      |
| Food from farm <sup>b</sup> | 138              | 0.78 (0.53-1.15)  | 0.88 (0.41-1.90) | 0.55 (0.29-1.05)    | 0.67 (0.46-0.98)                      |
| Hand dishwashing            | 126              | 0.49 (0.32-0.77)  | 0.21 (0.05-0.85) | 0.78 (0.42-1.42)    | 0.51 (0.34-0.77)                      |
| Home cooking <sup>c</sup>   |                  |                   |                  |                     |                                       |
| Never/almost never          | 72               | 1                 | 1                | 1                   | 1                                     |
| Approximately half the time | 533              | 0.88 (0.53-1.47)  | 1.67 (0.50-5.55) | 0.70 (0.36-1.34)    | 0.77 (0.47-1.26)                      |
| Most of the time/always     | 414              | 0.84 (0.51-1.40)  | 1.60 (0.47-5.44) | 0.55 (0.28-1.09)    | 0.72 (0.44-1.19)                      |
| Breastfeeding duration, mo  |                  |                   |                  |                     |                                       |
| 0-4                         | 145              | 1                 | 1                | 1                   | 1                                     |
| >4-8                        | 373              | 0.92 (0.62-1.36)  | 0.70 (0.34-1.41) | 0.86 (0.49-1.51)    | 0.87 (0.59-1.28)                      |
| $\geq 8$                    | 430              | 0.94 (0.64-1.39)  | 0.65 (0.33-1.31) | 0.99 (0.58-1.72)    | 0.90 (0.61-1.31)                      |

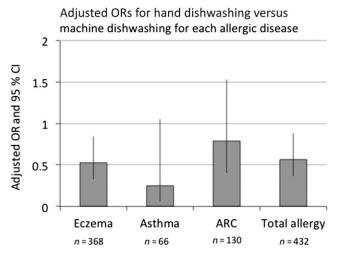
a Question: "Does the child eat food that includes fermented vegetables (such as sauerkraut or fermented cucumber) or other fermented foodstuffs?" Responses: never/almost never; at least once a month.

<sup>b</sup> Question: "Do you sometimes buy hens' eggs, meat, or unpasteurized milk directly from a farm?" Responses: yes; no.

<sup>c</sup> Question: "During the child's first year of life, how often was he/she given home-cooked food?" Responses: never/almost never; approximately half the time; most of the time/always.
<sup>d</sup> Any of eczema, asthma, or ARC.

not (P = .000). Traditional cooking might, however, be linked to different lifestyle factors in a similar way as hand dishwashing. We therefore adjusted for covariates and confounding variables with a backward multiple logistic regression-model as described in the Methods section (model 2). In the sixth and final step in the backward logistic regression-model, which included traditional cooking, father born in Sweden, day care attendance, parental history of allergy, and pet keeping during infancy, traditional cooking was still associated with a reduced risk of allergy (adjusted OR, 0.56; 95% CI: 0.41–0.77).

Even though eating fermented food and buying food from farms did not on their own reveal statistically significant associations with specific allergic diseases, a dose-response pattern was seen when analyzing these habits together with dishwashing habits. For this analysis,



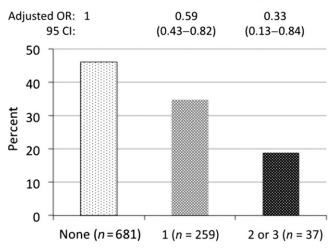
#### FIGURE 2

Adjusted ORs and 95% Cls for eczema, asthma, ARC, and total allergy (any of the 3 diseases) when analyzed with respect to dishwashing method used by the family. The prevalences of eczema and total allergy were significantly lower in children from families who use hand dishwashing (n = 126) instead of machine dishwashing (n = 868), whereas asthma revealed a borderline significance. Confounders were adjusted for in a hierarchical multiple logistic regression model, with parental history of allergy, day care attendance, and pet keeping during infancy as independent variables.

we choose total allergy as the outcome variable, and adjusted for confounders as previously described in the Methods section (model 2 and model 4). In families that used machine dishwashing, did not buy food directly from farms, and did not eat fermented food, allergic disease was reported by 46% of the children. The prevalence of allergic disease declined with increasing number of protective factors such that it was 19% for children with 2 or 3 protective factors (Fig 3).

#### **DISCUSSION**

The main finding from the current study is the association between allergy and common lifestyle factors, with lower allergy prevalence in children from families who mainly use hand dishwashing instead of machine dishwashing. This association was amplified if the children also ate fermented food or if the families bought food directly from farms. The importance of this finding is that in affluent countries there are common lifestyle factors with an individual impact that may affect the risk of allergy development in children. Several studies have previously revealed that living on a farm is associated with lower rates of atopic diseases.<sup>15</sup> However, such findings are mainly of interest from



#### Prevalence of total allergy in relation to number of traditional cooking factors

#### FIGURE 3

The prevalence of total allergy (eczema, asthma, or ARC) according to the number of protective factors reported for the child's family: that is, the number of factors included in the umbrella term traditional cooking (hand dishwashing, use of fermented food, or buying food from farm). Differences are expressed as adjusted ORs and 95% Cls, and confounders were adjusted for in a hierarchical multiple logistic regression model with parental history of allergy, day care attendance, pet keeping during infancy, and father born in Sweden, as independent variables.

a theoretical point of view only as living on a farm is not a practical recommendation for allergy prevention. Neither can the finding that allergy is less prevalent in several nonaffluent countries<sup>10</sup> be directly used in allergy-prevention measures, as the risks of living in an environment with low sanitary standards outweigh any possible benefits in terms of allergy protection. With this study, however, we have added a few further lifestyle factors to the list of those associated with protection against allergy that could be used in developed countries. The other factors on this list (ie, introducing fish into the child's diet at an early age,<sup>16</sup> having pets early in life,<sup>17</sup> and parental cleaning of the child's pacifier by sucking it)<sup>9</sup> are all lifestyle choices that could be applied by most people. Even though we do not currently have strong support for recommending any of these lifestyle factors in allergy prevention, they are already commonly used and most often regarded as harmless.

Low hygiene standards and increased microbial exposure are common

denominators often seen in low allergy-risk settings. Both a diverse gut microbial flora<sup>8</sup> and an altered oral microbial flora9 are associated with reduced rates of eczema. There is also an inverse relationship between allergy and a high bacterial content in drinking water,<sup>18</sup> and several studies have demonstrated that infections during childhood may prevent later development of allergy.<sup>6,19</sup> Factors related to the low allergy risk seen in many environments, such as in developing countries,<sup>10</sup> crowded living conditions,<sup>20</sup> early day care attendance,<sup>5</sup> having siblings,<sup>21</sup> and farm living,<sup>22</sup> are all supposed to be mediated via microbial immune stimulation in early life. Such a mechanism may also explain our findings. Fermentation of food is indeed a bacterial process, and it may also be assumed that hand dishwashing and buying food directly from farms are both associated with an increased microbial exposure compared with modern machine dishwashing and industrial processed food, respectively. The efficacy of

hand versus machine dishwashing was tested in 1000 New York restaurants in 1947.<sup>23</sup> Despite the use of machine dishwashers from the 1940s, they were more often superior to hand dishwashing in reducing bacterial content on utensils and cutlery. It is interesting to note the authors' remark in the discussion: "It is the considered belief of the present authors, as a result of the foregoing observations on hand dishwashing, that no great improvement in the quality of washed utensils will be obtained until a large majority of eating establishments have efficient mechanical or combination manualmechanical utensil washing devices." A more recent study from Bonn, Germany, confirms these results.<sup>24</sup> In this study, the authors compared different hand dishwashing techniques with modern machine dishwashers. They reported wide interindividual variations in dishwashing techniques and efficacy, and concluded that approximately half of the test persons did not achieve an acceptable result regarding the level of cleanliness. Furthermore, a person's hand dishwashing technique seemed to be important in reducing bacterial content. Lee et al<sup>25</sup> studied the efficiency of manual dishwashing conditions on bacterial survival on eating utensils. They concluded that efficacy is affected by the type of organic matter substrate, the shape of the utensil, the strength of the sanitizing agent, and the temperature of the washing water. In particular, milk products seemed to have the potential to remain on utensils at levels high enough to pose a health risk. Our assumption, as discussed above, that hand dishwashing will expose the family members to more bacteria than machine dishwashing is therefore plausible.

Fermented food and unpasteurized dairy products are often discussed in the contexts of farm living and the use of probiotics, and they will undoubtedly result in increased microbial exposure. Probiotics in particular have generated much research activity in recent years as they are thought to prevent several immune-mediated disorders. Even though there have been several interesting studies on probiotics in allergy prevention, the evidence is not yet strong enough to permit recommendations on the use of probiotics in allergy prevention.<sup>26</sup> The question is even more complicated when trying to isolate a possible allergy-preventive effect of unpasteurized dairy products. These products are mainly consumed by people living on farms with livestock; it is thus difficult to distinguish the allergy-preventing effect of general microbial exposure related to farm living per se from the bacterial exposure related to eating unpasteurized dairy products.<sup>27</sup>

When interpreting our results, it is wise to remember the observational study design. The inverse association between exposure and outcome is not necessary causal, and the mechanism discussed above is only 1 possible explanation. It is most likely that the lower prevalence of allergy seen in children from hand dishwashing families is related to lifestyle factors, but lifestyle factors other than dishwashing practices may be important. For example, overcrowded housing,<sup>20</sup> low socioeconomic status,<sup>28,29</sup> and immigration status<sup>30</sup> are all factors that have been associated with reduced allergy levels in various studies, and these factors may also be associated with dishwashing practices. We used multivariate OPLS analyses to screen the data for confounders and covariates, but despite adjusting for such factors in logistic regression models, the associations between hand dishwashing and allergy, and traditional cooking and allergy, remained, implying that a causal relationship might be possible.

Another limitation of the study is the cross-sectional, questionnaire-based

approach to measure exposure. Recall bias may be a problem, and we lack precise data on exposure. We cannot be absolutely sure of what the parents mean when they say that they use hand dishwashing; does it refer to current status or has it been their practice since the child was born? Similarly, the question on fermented food could be misunderstood, even though we gave examples of such foodstuffs, and we do not know exactly what type of food the families bought directly from farms. However, this type of weakness is a well-known limitation of the research method. When interpreting the results, the observational status of the study should be kept in mind, as should the fact that further studies are needed before any detailed conclusions can be made.

#### CONCLUSIONS

We found a lower prevalence of reported allergy in children aged 7 to 8 years from families who use hand dishwashing instead of machine dishwashing. This effect was further potentiated if they also ate fermented food or bought food directly from farms. We speculate that these lifestyle factors reduce allergy development via increased or more diverse microbial exposure, stimulating the immune system to develop in a more tolerant direction.

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