

childhood microbiota and food allergy in the FORWARD study; association with race and atopic phenotype.

Authors list

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## Introduction

In past decades there has been a sharp increase in the prevalence of all atopic conditions including food allergy (FA), asthma, allergic rhinitis and AD across the world. (1, 2) Following the original hygiene hypothesis in the 1980s, several studies have shown a link between changes in the environment and increases in atopic diseases. (3, 4) Studies provided evidence that a more limited exposure to microbes in early life can negatively impact the evolution of the human microbiota composition. The composition and diversity of the early life gut microbiome has been associated with development of atopic conditions. (5, 6) Furthermore, the presence of specific bacteria in the gut has been associated with protection against multiple allergic diseases including FA. (7-9) It has been recently shown that intestinal bacteria are critical for regulating allergic responses to dietary antigens, and protection against development of FA. (9)

Individuals of African origin who live in Western countries are at a significantly higher risk for allergic conditions and suffer from more severe allergic diseases than their White counterparts. (10-12) Black children with food allergy from US have higher rate of atopic comorbidities such as asthma and atopic dermatitis, FA-related adverse events and a different food allergen profile compared with white food allergic children. (12) It is not clear whether the gut microbiota plays any role in this differential phenotype.

Although previous microbiota studies have shown differences in the gut microbiota of children with and without FA, these differences have never been compared in children with from different races living within the same geographic area. Furthermore, to our knowledge it has never been determined whether these variations in the gut microbiome are linked to specific phenotypic severity traits among children with food allergy.

In the current study, we analysed and compared the stool microbiome of a group of black and white children aged 1-12 years recruited from four different cities in the US. We further sought to understand the impact of other clinical variables associated with FA phenotype comorbid allergic conditions, severity and rate of FA-related events and number of food allergens on the gut microbiome. Although all these clinical variables are regarded as important factors impacting how we treat and approach children with FA, currently there is no accepted method to measure the severity of FA. In this study we created a novel combined weighted index encompassing of these FA-related clinical variables to assess the association of gut microbiota with these factors in different races.

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## Methods

Subjects:

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Diet assessments:

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Assessment of phenotype and severity: All recruited children were assessed for comorbid asthma (or wheezing in younger children) and atopic dermatitis which was recorded in the initial survey. The types and number of food allergens that the child had reacted to and was tested positive for were captured through this survey. Furthermore, the parents were asked to

complete a survey about history of FA-related events and health care visits. These include FA-related anaphylaxis (captured through a specific multistep question assessing the systems involved during a reaction), need for auto injectable Epinephrine for treatment of FA-related event, emergency room visit and hospitalization for FA-related event. We created a weighted index from 8 carefully selected variables to capture the relative severity of the child's FA phenotype. (supplementary table 1)

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Stool collection: Parents were provided with stool specimen collection kits at the screening visit for baseline collection. Stool samples were collected with a 24-hour period, placed in sterile containers, and frozen immediately.

Microbiota analysis:

Microbiome statistical analysis:

## Results

## Discussion

## Conclusion

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