



Influence and effect of the human microbiome in allergy and asthma

Ariane R. Panzer and Susan V. Lynch

Purpose of review

Studies have illustrated that the healthy human microbiome (i.e. the communities of microbes, their genomic content and interaction with the host) plays a role in the maintenance of immune homeostasis. Perturbation of these communities is an emerging characteristic of an increasing number of inflammatory diseases. The goal of this article is to review the current literature on both respiratory and gut microbiomes and their established relationship with allergy and asthma.

Recent findings

Multiple studies have demonstrated airway microbiota dysbiosis, characterized by *Proteobacteria* expansion in the lower airways, to be a consistent trait of established adult asthma. Members of this phylum are associated with disease features such as bronchial hyperreactivity or corticosteroid resistance. Emerging evidence implicates the neonatal gut microbiome as playing a significant role in the development of childhood atopy, a common precursor to asthma. Murine studies have demonstrated that specific bacterial species (e.g. *Lactobacillus johnsonii*, *Bacteroides fragilis*) and microbial metabolites (e.g. the short-chain fatty acid propionate), when supplemented to animals, confer protection against allergen-induced airway disorders.

Summary

The emerging view of atopy and asthma is one consistently related to inappropriate microbial community composition and function in both the airway and gastrointestinal tract. This opens up the possibility that strategies to rationally manipulate microbiota at these sites may represent a novel approach to disease prevention or management.

Keywords

airway, allergy, asthma, gut, microbiota

INTRODUCTION

The hypothesis that the microbes residing in and on the human body play a role in dictating health status dates back to Louis Pasteur [1]. However, the recent advent of molecular methods to identify and classify microorganisms from mixed-species microbial communities in a culture-independent manner has dramatically expanded our view of the diversity of microbes that colonize the human host. Microbiota (a biomarker gene-based catalog of the microorganisms found in a given ecosystem) profiling approaches most commonly utilize the 16S ribosomal RNA (16S rRNA) gene as a biomarker for molecular profiling of bacteria and archaea present. This gene is ubiquitous among the members of these kingdoms, but is not encoded by higher organisms, and hence provides a useful biomarker gene for exclusively examining members of these microbial kingdoms. The 16S rRNA gene encodes regions of sequence conservation, which flank nine

hypervariable sequence regions (V1–V9), permitting PCR amplification of either the entire gene or selected hypervariable regions. The sequence of the hypervariable region(s) permits classification of organisms into phylogenetically related groups, or operational taxonomic units, based on operator-defined sequence similarity of the PCR-amplified region. Sequencing a 16S rRNA amplicon pool generated from DNA extracted from a mixed-species microbial community enables identification, phylogeny enumeration (community richness) as well as a semi-quantitative assessment of operational

Division of Gastroenterology, Department of Medicine, University of California San Francisco, San Francisco, California, USA

Corresponding to Susan V. Lynch, PhD, University of California San Francisco, Division of Gastroenterology, San Francisco, CA 94143, USA. Tel: +1 415 476 6784; e-mail: susan.lynch@ucsf.edu

Curr Opin Rheumatol 2015, 27:373–380

DOI:10.1097/BOR.0000000000000191

KEY POINTS

- Compositional differences in airway microbial communities are associated with features of established adult asthma such as bronchial hyperactivity and corticosteroid resistance.
- Neonatal gastrointestinal microbial community composition is related to childhood allergy susceptibility.
- Many early-life exposures associated with allergy and asthma have been shown to influence respiratory and gastrointestinal microbial communities.
- Murine studies have demonstrated that gut microbiome composition and metabolic function are associated with allergic response in the airways.

taxonomic unit abundance and their distribution (evenness) within and across microbial communities. A workflow for this process is provided in Fig. 1.

Recent technological advances, specifically the development of high-throughput next-generation sequencing platforms, has decreased costs associated with sequencing. This has increased read depth and community coverage, allowing identification of dominant and rarer organisms in these assemblages. In addition to examining community membership, recent efforts have focused on understanding microbial community function. Shotgun sequencing of DNA extracted from a microbial community provides information on gene content and functional capacity of the assemblage (Metagenomics), whereas a similar shotgun sequencing approach applied to cDNA generated from mRNA extracts permits assessment of the community-wide transcriptional profile (Metatranscriptomics). Beyond

advances in sequence technology, innovations in mass spectroscopy permit global profiling of protein expression (Metaproteomics) or metabolic (Metabolomic) activity of the microbiome. Currently efforts in the field are focused on integrating such 'multiomic' approaches in an attempt to provide a systems biology strategy to understand not only microbial community composition but also the functional output of these assemblages and their interaction with the human host. It is anticipated that exploring microbial communities from this perspective will offer a richer understanding of human–microbial relationships and reshape our understanding of human health and disease.

The magnitude of the microbiome's contribution to human health is underscored by the sheer burden of microorganisms that colonize humans; (currently estimated to outnumber human cells by 3:1) [2]. The genetic capacity of the healthy microbiome, which is estimated to house up to several thousand distinct species [3], encodes approximately 2 000 000 genes, 100 times that of the human genome [4]. In healthy individuals, this ancillary panmicrobial genome provides a wealth of essential functions and indicates a long and largely symbiotic coevolution of man and microbes. Essential functions encoded by the microbiome have largely been determined in the gut microbiome, where the capacity to digest and metabolize complex carbohydrates and biosynthesize essential vitamins such as vitamin B12 are functions exclusively encoded by our microbial inhabitants [5]. An additional critical influence of the microbiome is its capacity to modulate both innate and adaptive host immune responses [6]. Given the emerging role of T-cell subset proliferation in a range of chronic inflammatory and autoimmune diseases, for example, asthma

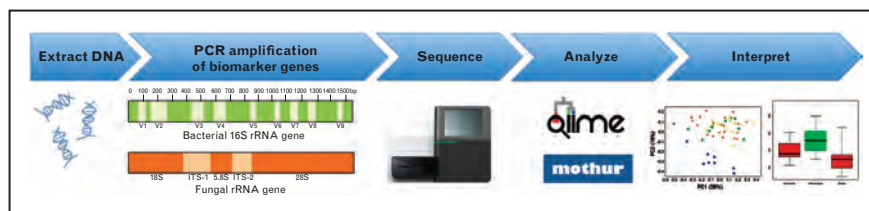


FIGURE 1. Workflow for profiling microbiota. DNA is extracted from samples followed by PCR amplification of specific biomarker genes. For bacterial community composition profiling, the 16S rRNA gene is amplified with specific PCR primers targeting one or more consecutive hypervariable regions. For mycological characterization the Internal transcribed spacer region 2 (ITS-2) of the fungal rRNA gene is commonly amplified. Following amplification, samples are purified, pooled, and sequenced. Sequencing data are then analyzed using pipelines such as Quantitative Insights Into Microbial Ecology or Mothur, which support a variety of data analyses tools. Findings are frequently interpreted in terms of α (intra) or β (inter) diversity. For example, summary statistics of α diversity are commonly used that enumerate phylotypes (community richness), determine their distribution within a community (evenness), and assess their diversity. β diversity enables assessment of community dissimilarity across samples within a given sample set, typically visualized by principal components or coordinates analyses. These approaches may also be used in permutational multivariate testing, to examine relationships between microbial community composition and various factors such as clinical data.

(Th2; [7]), rheumatoid arthritis, systemic lupus erythematosus, and inflammatory bowel disease (Th17; reviewed in [8]), the influence of the microbiome on the development of T-cell populations has become a major focus. Seminal work by Ivanov *et al.* [9] demonstrated that segmented filamentous bacteria, identified by comparative microbiota profiling, are highly enriched in mice with large numbers of ileal Th17 cells. Furthermore, oral gavage of fecal material containing segmented filamentous bacteria induced ileal Th17 proliferation in previously deficient animals. In independent studies, Mazmanian *et al.* [10] demonstrated that capsular polysaccharide A, a repeating four-sugar polymer produced on the surface of *Bacteroides fragilis*, promotes production of T-helper 1 (Th1) cytokines. This same polymer was also recently shown, in an independent study [11], to induce interleukin-10-producing regulatory T cells via toll-like receptor 2-dependent plasmacytoid dendritic cell activation. Similarly, gut microbial species belonging to *Clostridia* clades IV and XIV have been shown in mice to induce T-regulatory cells, which downregulate proinflammatory adaptive responses [12], including those associated with allergic asthma. Although still largely focused on single species or phylogenetically related groups of bacteria, these findings highlight the influence members of the gut microbiome exert on dictating the tone of host immune response. This opens up the possibility that microbial dysbioses and loss of species and functions critical to induction of immune homeostasis may be responsible for a range of human inflammatory diseases whose etiology is currently unknown.

ASTHMA AND THE AIRWAY MICROBIOME

One such disease of hyperactive adaptive immunity is allergic asthma. As of 2010, approximately 25 million Americans were diagnosed with asthma, which incurs \$56 billion per year in related healthcare costs in the United States [13]. This healthcare expenditure, in conjunction with the steady increase in the prevalence during the last several decades, particularly among pediatric populations in Western nations, makes allergic asthma one of the most common and serious chronic inflammatory diseases [14]. Asthma is classically considered as an overactivation of the Th2 arm of adaptive immunity characterized by increased expression of Th2 proinflammatory cytokines interleukin-4, interleukin-5, and interleukin-13, increased serum immunoglobulin E antibodies and increased numbers of airway eosinophils and mast cells. However, this view of the disease is now evolving to encompass subgroups of patients who do not exhibit Th2

overactivation, but are rather characterized by involvement of distinct arms of adaptive immunity, for example, smoker-associated Th1 hyperactivation [15] or asthmatic patients who exhibit aberrant Th17 proliferation [16].

Although the upper airways of healthy individuals are robustly colonized by a diversity of bacterial species [17], the lower airways possess a significantly smaller bacterial burden [18]. Culture-independent studies of asthmatic patients and healthy individuals have demonstrated that asthmatics possess both a higher burden of bacteria in their lower airways, as well as a significantly increased diversity of microbes when compared with healthy individuals [18]. Despite distinct patient populations and microbiota profiling platforms, all of the studies performed to date have demonstrated a characteristic expansion of organisms belonging to the *Proteobacteria* in asthmatics, implicating members and activities of these species in the pathogenesis of disease. Indeed a study by Huang *et al.* [18], who examined the airway microbiota of asthmatic patients using inhaled corticosteroids ($n = 65$), demonstrated that the degree of bronchial hyperresponsiveness, a measure of disease severity, was significantly related to the composition of the lower airway microbiota composition. More specifically, members of the *Comamonadaceae*, *Sphingomonadaceae*, and *Oxalobacteraceae*, all belonging to the phylum *Proteobacteria*, were among those organisms exhibiting the strongest correlations with airway hyperresponsiveness. Other studies have demonstrated that disease phenotypes, such as corticosteroid resistance among asthmatic individuals, are related to differences in airway microbiota composition [19]. Goleva *et al.* also examined bronchoalveolar lavage of 51 individuals, including 39 asthmatic individuals, 29 of whom exhibited corticosteroid resistance. Corticosteroid resistant patients exhibited expansion of *Haemophilus parainfluenza*, a member of the *Proteobacteria*. The investigators demonstrated using *in vitro* studies that this species induced p38 mitogen-activated protein kinase (MAPK) activation, increased interleukin-8 ($P < 0.01$), mitogen-activated kinase phosphatase 1 mRNA ($P < 0.01$) expression, and inhibition of corticosteroid responses ($P < 0.05$) in asthmatic airway macrophages. These phenotypes could be overcome by inhibiting transforming growth factor- β -associated kinase-1 (TAK1; an upstream activator of MAPK). Hence, the emerging data indicate that asthmatic airways exhibit an aberrant lower airway microbiota, characterized by an expansion of members of the *Proteobacteria*, and that particular disease features, such as bronchial hyperreactivity or corticosteroid responsiveness, are related to the

presence and activities of a subset of organisms within the Proteobacteria.

EARLY-LIFE AIRWAY MICROBIOME AND ALLERGIC ASTHMA

Although the aforementioned studies primarily focus on adult asthma, an increasing number of investigations implicate the neonatal microbiome in the subsequent development of childhood atopy. Although atopic children do not always develop asthma, recent data have indicated that allergic children who exhibit multiple early-life (age 2) sensitizations to a range of allergens, for example, dog, cat, cockroach, egg, peanut, are at a significantly higher risk for subsequently developing physician diagnosed asthma at age 4 (odds ratio, 5.3; 95% confidence interval, 1.6–17.4), compared with those who are either nonsensitized or sensitized to only one or two of these allergens [20]. Indeed, murine studies examining neonatal mice whose airways were sensitized with house dust mite (HDM) demonstrated more pronounced allergic airway inflammation compared with similarly challenged adult animals [21[¶]]. The diminished allergic response to HDM in adult animals was related to age-dependent changes in bacterial load and community composition in the airways and to the promotion of anti-inflammatory T-regulatory cells. Furthermore, the absence of microbial colonization during the first 2 weeks of life exaggerated neonatal allergic reaction to HDM, which persisted even in adulthood, thereby supporting the importance of early-life microbial colonization for protection from more severe atopy development.

Early-life recurrent wheeze is commonly (though not always) a precursor to asthma development in childhood. Unlike asthma, however, recurrent wheeze is immunologically characterized by nonspecific inflammation dominated not by eosinophils (as is typically the case in asthmatic airways) but rather by macrophages and neutrophils, cell types associated with response to an infectious agent [22]. This has led to the hypothesis that early-life infection, or perhaps response to aberrant microbial colonization of the airways with pathogenic members, may represent a common predecessor to asthma development in childhood. Bisgaard *et al.* [23] examined hypopharyngeal aspirates from infants at high risk for developing asthma using culture-based methods and demonstrated that overt colonization by *Streptococcus pneumoniae*, *Haemophilus influenzae*, *Moraxella catarrhalis*, or a combination of these organisms at 1 month of age was associated with an increased risk of asthma development by age 5. Importantly, the detection of these species at

12 months of age was not associated with the same risk of disease development, pointing to the importance of appropriate early-life microbiome development in influencing subsequent allergic immune responses.

It should be noted that children who experience a severe response to respiratory infection typically requiring hospitalization are at a significantly higher risk for developing childhood asthma [24]. Clues as to how enrichment of these specific bacterial species in the airways may influence subsequent disease development has emerged from a recent study [25] of nasal samples of 308 children with and without asthma who were examined for the presence of *S. pneumoniae*, *H. influenzae*, and *M. catarrhalis* during the peak rhinovirus season. The authors found that rhinovirus infection was associated with increased detection of these bacterial species and that the presence of either *M. catarrhalis* or *S. pneumoniae* in conjunction with rhinovirus significantly increased cold and asthma symptoms [25]. Hence, these data indicate that the increased abundance of specific respiratory pathogens serves to potentiate the severity of viral respiratory infection and thus risk of subsequent asthma development.

ROLE OF THE GUT MICROBIOME IN ALLERGIC ASTHMA

In addition to airway microbial communities playing a critical role in asthma, there is growing evidence that differences in gut microbial communities influence systemic immune function and thus contribute to the development of disease at sites remote from the gastrointestinal tract, including allergic airway disease (Table 1) [26–28,29[¶],30,31[¶]]. Studies by Penders *et al.* [26], and, independently by Kalliomaki *et al.* [27] have demonstrated, using culture-based approaches, that detection of high abundances of either *Escherichia coli* or *Clostridium difficile* in neonatal feces is associated with significantly higher risk of developing childhood allergic disease. In parallel with species overgrowth, the absence of specific species such as *Lactobacillus* or *Bifidobacterium* in neonatal stool has also been associated with an increased risk of childhood atopy development [28]. Hence, the developing hypothesis is that, like the airway, aberrant microbiome development in the neonatal gut lays the foundation for allergic disease and subsequently asthma development in childhood.

The gut microbiome arguably represents the best-studied microbiome in the human superorganism. Although recent studies have described the presence of a placental-associated microbiome in

Table 1. Selection of studies supporting the role of the gut microbiome in airway health

Organism(s)	Model	Mechanism	Influence	Sources
<i>Escherichia coli</i> ; <i>Clostridia</i>	Human	Unknown	Increased risk of atopy	[26,27]
<i>Lactobacillus</i> ; <i>Bifidobacterium</i>	Human	Unknown	Decreased risk of atopy	[28]
<i>Lactobacillus johnsonii</i>	Murine	Unknown	Reduced airway Th2 cytokines	[29 [■]]
<i>Bacteroides fragilis</i>	Murine	Polysaccharide A	FoxP3(+) regulatory T-cell prevention of Th1 and Th2 responses	[30]
<i>Bacteroidaceae</i> ; <i>Bifidobacteriaceae</i>	Murine	Short-chain fatty acid production	Increased hematopoiesis of dendritic cell precursors, dampened Th2 response	[31 [■]]

full-term deliveries [32], the birthing process appears to represent a strong influence on the microbiome present in neonates. Vaginal or Caesarian delivery results in distinct microbiota present in the oral cavity, skin, and gut of neonates. The former is characterized by an abundance of genera such as *Lactobacillus* and *Sneathia* found in the mother's vaginal tract, and the latter by characteristic skin-associated organisms such as *Staphylococcus* and *Streptococcus* [33]. Importantly, infants delivered via Cesarean section have an increased risk of atopy compared with vaginally delivered infants [34], supporting the hypothesis that early-life patterns of microbial colonization and microbiome development play a role in childhood allergic disease development. Ecologically, initial colonizers, or pioneering organisms, frequently determine ecosystem conditions and in-so-doing dictate the pattern of co-colonizing organisms [35]. The gut microbiome develops and increases in diversity during the first several years of life, until between 1 and 3 years when it resembles that of an adult assemblage with respect to bacterial phylogenetic distributions [35]. Hence early-life experiences, such as birthing method, neonatal nutrition, antimicrobial administration, and environmental microbial exposures among others, that are known to influence the composition of the gut microbiome, may significantly influence childhood allergic disease susceptibility.

Indeed, multiple independently validated risk factors for childhood allergic asthma are centered on this critical period of early-life development. Antimicrobial exposure during early infancy increases the risk of childhood atopy [36] as well as asthma, rhinoconjunctivitis, and eczema [37]. Conversely, interaction with other children either via siblings at home or through attendance of day-care is associated with protection against asthma and wheeze [38]. Additionally, environmental exposures during pregnancy can impact the risk of disease development in offspring. Infants whose mothers were administered antimicrobials during the third trimester of pregnancy had an increased risk of asthma exacerbation, whereas infants whose

mothers received antimicrobials at any point during pregnancy had an increased risk of corticosteroid use and asthma-related hospitalization [39]. Studies have also considered the influences of furred pet exposure during pregnancy and show that infants had lower cord blood immunoglobulin E levels (the antibody response classically elevated in allergic disease) when their mothers resided with either cats or dogs in the home during pregnancy [40].

Diet represents a strong selective pressure on the gut microbiome [41,42]. Unsurprisingly, therefore, mode of nutrition, particularly early-life formula feeding, represents a significant risk factor for childhood allergic disease. A study [43] comparing microbial constituents present in fecal samples of breast-fed or formula-fed infants found that the dominant members of breast-fed infant microbiota belonged to the *Bifidobacteria* and that *Lactobacillus* and *Streptococcus* species, fermentative species that produce anti-inflammatory short-chain fatty acids (SCFAs), were also enriched in these infants. Formula-fed infants possessed a compositionally distinct microbiome, typically co-dominated by *Bacteroides* and *Bifidobacteria* and also enriched for *Staphylococcus*, *Escherichia*, and *Clostridium* species [43]. Hence, the emerging view is that early-life exposures that influence the composition, and therefore the function of the gut microbiome and its capacity to appropriately educate the maturing host immune response, are associated with childhood atopy (Fig. 2).

The depletion of key commensal species from these microbial assemblages appears a common and pervasive characteristic of infants at high risk of disease development. Evidence that commensal gut microbes play a critical role in modulating airway immune responses comes from several studies, including those demonstrating that antimicrobial depletion of the gut microbiome leads to enhanced airway pathogenicity in response to viral respiratory infection [44]. Fujimura *et al.* [29[■]] recently demonstrated that commensal *Lactobacillus johnsonii* supplementation of mice protected animals against cockroach allergen or ovalbumin

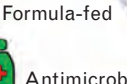
Factors	Organisms	Influence
 Vaginal delivery  Breast-fed  Exposure to animals	<i>Lactobacillus</i> & <i>Sneathia</i>  <i>Bifidobacteria</i> & <i>Lactobacillus</i>  <i>Lactobacillus</i> <i>johnsonii</i>	DECREASED risk of allergy and asthma 
 Cesarean section  Formula-fed  Antimicrobials  Urban environment	<i>Staphylococcus</i> & <i>Streptococcus</i>  <i>Bacteroides</i> & <i>Bifidobacteria</i> <i>Staphylococcus</i> , <i>E.coli</i> , <i>Clostridia</i>  Reduced <i>Firmicutes</i> & <i>Bacteroidetes</i>	INCREASED risk of allergy and asthma 

FIGURE 2. Early-life factors influencing microbial community members and risk of allergy and asthma. Many factors play a role in shaping human microbial communities. Infants who are delivered vaginally, who are breast-fed, and/or who are exposed to furred animals are characterized by an enrichment of *Lactobacillus* and *Sneathia*, *Bifidobacteria* and *Lactobacillus*, and *Lactobacillus johnsonii*, respectively. Infants who are exposed to these environmental factors have a decreased risk of developing allergy and asthma. Infants delivered via Cesarean section, who are formula-fed, and/or who live in urban environments are characterized by an enrichment of *Staphylococcus* and *Streptococcus*, *Bacteroidetes* and *Bifidobacteria* along with *Staphylococcus*, *Escherichia coli*, and *Clostridia*, and a depletion of *Firmicutes* and *Bacteroidetes*, respectively. Infants exposed to these environmental factors, as well as early-life antimicrobial exposure, have an increased risk of developing allergy and asthma.

airway sensitization. Protection was associated with significantly downregulated Th2 cytokine (interleukin-4, interleukin-5, and interleukin-13) expression in the airways. Supplementation with this species also protected mice against respiratory syncytial virus infection. Protected mice exhibited the capacity to mount a normal interferon- γ response (necessary for viral clearance), but significantly reduced Th2 and Th17 airway responses. Protected animals possessed a significantly restructured gut microbiome composition and reduced numbers of activated dendritic cells in their mesenteric lymph nodes, suggesting that microbial-mediated alterations to the antigen-presenting cell population represents at least one mechanism of protection against both viral and allergen challenge. Independently, a recent study [30] demonstrated that gastrointestinal exposure to polysaccharide A from *B. fragilis*, prevented Th1 and Th2 responses via noncanonical FoxP3(+) regulatory T cells which produced interleukin-10.

Additional mechanisms by which the gut microbiome may influence both local and remote

mucosal responses is via immunomodulatory metabolites. Trompette *et al.* [31] recently demonstrated that mice fed a high-fiber diet exhibited significantly decreased airway allergic response, characterized by reduced interleukin-4, interleukin-5, interleukin-13, and interleukin-17A expression in lung tissue, following intranasal sensitization with HDM (compared with isogenic animals fed a low-fiber diet). Indeed, in mice consuming a high-fiber diet enriched for members of the *Bacteroidaceae* and *Bifidobacteriaceae* in the gut microbiome, including members capable of fermenting fiber to anti-inflammatory SCFAs, an increase in production and circulation of SCFA was observed. Subsequent studies involving supplementation of animals with the SCFA, propionate, also led to a decreased susceptibility to allergic airway inflammation and increased hematopoiesis of dendritic cell precursors resulting in lung dendritic cells that were less effective at reactivating Th2 responses. Collectively these data, while representing proof-of-principle studies in murine models, suggest that targeting gut microbial communities,

via purposeful supplementation strategies coupled with dietary interventions that shape gut microbiome composition and function may represent a major tool in preventing, treating, or managing allergic asthma.

CONCLUSION

Many factors influence asthma and allergy development including early-life environmental exposures or lack thereof. Several of these exposures are established influences on, or are plausibly linked to, shaping microbiome composition at both the respiratory and gastrointestinal mucosal surfaces. Because microbial species sense and respond to each other and, more specifically, the quantity of other species in their local environment, these compositional changes engender functional changes in microbiome output and altered host interactions. The emerging evidence indicates that these functional changes influence host immune responses in a manner that dictates disease susceptibility. Studies have indicated that both airway and gastrointestinal microbiomes contribute to the development of allergy and asthma, and that microbial metabolic products serve to protect the airways. Thus, gut microbiome manipulation in very early life when these communities are developing phylogenetic diversity and functional complexity in the context of a developing immune response may offer a novel opportunity to intervene and change the course of disease.

Acknowledgements

None.

Financial support and sponsorship

S.V.L.'s asthma microbiome research program is supported by The National Institutes of Health awards from the NIAID: AI089473-01, AI113916 and NHLBI: U10 HL098107-05.

Conflicts of interest

There are no conflicts of interest.

REFERENCES AND RECOMMENDED READING

Papers of particular interest, published within the annual period of review, have been highlighted as:

- of special interest
- of outstanding interest

1. Mackowiak PA. The normal microbial flora. *N Engl J Med* 1982; 307:83–93.
2. Savage DC. Microbial ecology of the gastrointestinal tract. *Annu Rev Microbiol* 1977; 31:107–133.
3. Rajilic-Stojanovic M, de Vos WM. The first 1000 cultured species of the human gastrointestinal microbiota. *FEMS Microbiol Rev* 2014; 38:996–1047.
4. Ley RE, Peterson DA, Gordon JL. Ecological and evolutionary forces shaping microbial diversity in the human intestine. *Cell* 2006; 124:837–848.
5. Yatsunenko T, Rey FE, Manary MJ, *et al.* Human gut microbiome viewed across age and geography. *Nature* 2012; 486:222–227.
6. Jarchum I, Pamer EG. Regulation of innate and adaptive immunity by the commensal microbiota. *Curr Opin Immunol* 2011; 23:353–360.
7. Woodruff PG, Modrek B, Choy DF, *et al.* T-helper type 2-driven inflammation defines major subphenotypes of asthma. *Am J Respir Crit Care Med* 2009; 180:388–395.
8. Han L, Yang J, Wang X, *et al.* Th17 cells in autoimmune diseases. *Front Med* 2015; 9:10–19.
9. Ivanov II, Atarashi K, Manel N, *et al.* Induction of intestinal Th17 cells by segmented filamentous bacteria. *Cell* 2009; 139:485–498.
10. Mazmanian SK, Liu CH, Tzianabos AO, *et al.* An immunomodulatory molecule of symbiotic bacteria directs maturation of the host immune system. *Cell* 2005; 122:107–118.
11. Dasgupta S, Erturk-Hasdemir D, Ochoa-Reparaz J, *et al.* Plasmacytoid dendritic cells mediate anti-inflammatory responses to a gut commensal molecule via both innate and adaptive mechanisms. *Cell Host Microb* 2014; 15:413–423.
12. Atarashi K, Tanoue T, Shima T, *et al.* Induction of colonic regulatory T cells by indigenous *Clostridium* species. *Science* 2011; 331:337–341.
13. Vital Signs: Asthma in the U.S. Growing Every Year. <http://www.cdc.gov/VitalSigns/pdf/2011-05-vitalsigns.pdf>. U.S. Centers for Disease Control and Prevention; 2011. [Accessed May, 2011]
14. Akinbami LJ, Moorman JE, Bailey C, *et al.* Trends in asthma prevalence, healthcare use, and mortality in the United States, 2001–2010. *NCHS Data Brief* 2012; (94):1–8.
15. Thomson NC, Chaudhuri R, Livingston E. Asthma and cigarette smoking. *Eur Respir J* 2004; 24:822–833.
16. Wong CK, Lun SW, Ko FW, *et al.* Activation of peripheral Th17 lymphocytes in patients with asthma. *Immunol Invest* 2009; 38:652–664.
17. Abreu NA, Nagalingam NA, Song Y, *et al.* Sinus microbiome diversity depletion and *Corynebacterium tuberculoostearicum* enrichment mediates rhinosinusitis. *Sci Transl Med* 2012; 4:151ra24.
18. Huang YJ, Nelson CE, Brodie EL, *et al.* Airway microbiota and bronchial hyperresponsiveness in patients with suboptimally controlled asthma. *J Allergy Clin Immunol* 2011; 127:372–381; e1–3.
19. Goleva E, Jackson LP, Harris JK, *et al.* The effects of airway microbiome on corticosteroid responsiveness in asthma. *Am J Respir Crit Care Med* 2013; 188:1193–1201.
20. Havstad S, Johnson CC, Kim H, *et al.* Atopic phenotypes identified with latent class analyses at age 2 years. *J Allergy Clin Immunol* 2014; 134:722–727; e2.
21. Gollwitzer ES, Saglani S, Trompette A, *et al.* Lung microbiota promotes tolerance to allergens in neonates via PD-L1. *Nat Med* 2014; 20:642–647. Demonstrated using a murine model that microbial exposure during the critical neonatal period dictates responsiveness to aeroallergens in adulthood.
22. Bisgaard H. Persistent wheezing in very young preschool children reflects lower respiratory inflammation. *Am J Respir Crit Care Med* 2001; 163:1290–1291.
23. Bisgaard H, Hermansen MN, Buchvald F, *et al.* Childhood asthma after bacterial colonization of the airway in neonates. *N Engl J Med* 2007; 357:1487–1495.
24. Johnston SL, Pattemore PK, Sanderson G, *et al.* The relationship between upper respiratory infections and hospital admissions for asthma: a time-trend analysis. *Am J Respir Crit Care Med* 1996; 154 (3 Pt 1):654–660.
25. Kloefer KM, Lee WM, Pappas TE, *et al.* Detection of pathogenic bacteria during rhinovirus infection is associated with increased respiratory symptoms and asthma exacerbations. *J Allergy Clin Immunol* 2014; 133:1301–1307; 7 e1–3.
26. Penders J, Stobberingh EE, Thijs C, *et al.* Molecular fingerprinting of the intestinal microbiota of infants in whom atopic eczema was or was not developing. *Clin Exp Allergy* 2006; 36:1602–1608.
27. Kalliomaki M, Kirjavainen P, Eerola E, *et al.* Distinct patterns of neonatal gut microflora in infants in whom atopy was and was not developing. *J Allergy Clin Immunol* 2001; 107:129–134.
28. Sjogren YM, Jenmalm MC, Bottcher MF, *et al.* Altered early infant gut microbiota in children developing allergy up to 5 years of age. *Clin Exp Allergy* 2009; 39:518–526.
29. Fujimura KE, Demoor T, Rauch M, *et al.* House dust exposure mediates gut microbiome *Lactobacillus* enrichment and airway immune defense against allergens and virus infection. *Proc Natl Acad Sci U S A* 2014; 111:805–810. Demonstrated that environmental house dust exposure impacts the gut microbiome in a manner that can protect against airway allergy challenge and respiratory viral infection.
30. Johnson JL, Jones MB, Cobb BA. Bacterial capsular polysaccharide prevents the onset of asthma through T-cell activation. *Glycobiology* 2015; 25:368–375.
31. Trompette A, Gollwitzer ES, Yadava K, *et al.* Gut microbiota metabolism of dietary fiber influences allergic airway disease and hematopoiesis. *Nat Med* 2014; 20:159–166.

An important study showing that diet can influence microbial community composition and function and that diet-altered communities can modulate immune responses to allergic inflammation through production of anti-inflammatory short-chain fatty acids.

32. Aagaard K, Ma J, Antony KM, *et al.* The placenta harbors a unique microbiome. *Sci Transl Med* 2014; 6:237ra65.
33. Dominguez-Bello MG, Costello EK, Contreras M, *et al.* Delivery mode shapes the acquisition and structure of the initial microbiota across multiple body habitats in newborns. *Proc Natl Acad Sci U S A* 2010; 107:11971–11975.
34. Renz-Polster H, David MR, Buist AS, *et al.* Caesarean section delivery and the risk of allergic disorders in childhood. *Clin Exp Allergy* 2005; 35:1466–1472.
35. Palmer C, Bik EM, DiGiulio DB, *et al.* Development of the human infant intestinal microbiota. *PLoS Biol* 2007; 5:e177.
36. Johnson CC, Ownby DR, Alford SH, *et al.* Antibiotic exposure in early infancy and risk for childhood atopy. *J Allergy Clin Immunol* 2005; 115:1218–1224.
37. Foliaki S, Pearce N, Bjorksten B, *et al.* Antibiotic use in infancy and symptoms of asthma, rhinoconjunctivitis, and eczema in children 6 and 7 years old: International Study of Asthma and Allergies in Childhood Phase III. *J Allergy Clin Immunol* 2009; 124:982–989.
38. Ball TM, Castro-Rodriguez JA, Griffith KA, *et al.* Siblings, day-care attendance, and the risk of asthma and wheezing during childhood. *N Engl J Med* 2000; 343:538–543.
39. Stensballe LG, Simonsen J, Jensen SM, *et al.* Use of antibiotics during pregnancy increases the risk of asthma in early childhood. *J Pediatr* 2013; 162:832–838; e3.
40. Aichbhaumik N, Zoratti EM, Strickler R, *et al.* Prenatal exposure to household pets influences fetal immunoglobulin E production. *Clin Exp Allergy* 2008; 38:1787–1794.
41. Carmody RN, Gerber GK, Luevano JM Jr, *et al.* Diet dominates host genotype in shaping the murine gut microbiota. *Cell Host Microbe* 2015; 17:72–84.
42. David LA, Maurice CF, Carmody RN, *et al.* Diet rapidly and reproducibly alters the human gut microbiome. *Nature* 2014; 505:559–563.
43. Harmsen HJ, Wildeboer-Veloo AC, Raangs GC, *et al.* Analysis of intestinal flora development in breast-fed and formula-fed infants by using molecular identification and detection methods. *J Pediatr Gastroenterol Nutr* 2000; 30:61–67.
44. Ichinohe T, Pang IK, Kumamoto Y, *et al.* Microbiota regulates immune defense against respiratory tract influenza A virus infection. *Proc Natl Acad Sci U S A* 2011; 108:5354–5359.