



Contents lists available at ScienceDirect

Allergy International

journal homepage: <http://www.elsevier.com/locate/alit>

Invited review article

Development of the gut microbiota in infancy and its impact on health in later life

Masaru Tanaka, Jiro Nakayama*

Department of Bioscience and Biotechnology, Faculty of Agriculture, Graduate School, Kyushu University, Fukuoka, Japan

ARTICLE INFO

Article history:

Received 8 June 2017

Received in revised form

12 July 2017

Accepted 14 July 2017

Available online xxx

Keywords:

Allergy

Bifidobacterium

Gut microbiota

Hygiene hypothesis

Infants

Abbreviations:

GI, gastrointestinal; IBD, inflammatory bowel disease; SCFA, short chain fatty acids; Treg, regulatory T; Th, helper T; Ig, immune globulin; HMO, oligosaccharides in human milk; GOS, galactooligosaccharide; IL, interleukin; FA, fatty acids; SFB, segmented filamentous bacteria; Foxp3, Forkhead box P3; tTreg, Treg in the thymus; pTreg, Treg in the periphery; ATP, adenosine triphosphate

ABSTRACT

Gut microbial ecology and function are dynamic in infancy, but are stabilized in childhood. The 'new friends' have a great impact on the development of the digestive tract and host immune system. In the first year of life, especially, the gut microbiota dramatically changes through interactions with the developing immune system in the gut. The process of establishing the gut microbiota is affected by various environmental factors, with the potential to be a main determinant of life-long health. In this review, we summarize recent findings regarding gut microbiota establishment, including the importance of various factors related to the development of the immune system and allergic diseases later in life.

Copyright © 2017, Japanese Society of Allergology. Production and hosting by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

Introduction

Several hundred bacterial species and a total of 10^{14} cells colonize the human gastrointestinal (GI) tract in a mutualistic relationship with the host and its immune system.^{1,2} A healthy gut microbiota is stable and serves various useful functions such as metabolizing barely digestible polysaccharides,^{3,4} detoxifying toxic products,⁵ serving as a barrier against pathogens,^{6,7} and aiding in the development of the host immune system.⁸ However, it has become clear through recent studies that dysbiosis, in which the symbiotic relationship between the host and gut microbiota is altered, is associated with various diseases, including inflammatory

bowel disease (IBD),⁹ irritable bowel syndrome,¹⁰ obesity,^{11–14} allergy,¹⁵ autoimmune disease, and brain disorders.¹⁶

Although intestinal bacterial colonization begins when a fetus is in the lower uterus, an infant's gut microbiota is established after birth. The establishment of a stable gut microbiota generally accompanies two big transitions in infancy. The first transition occurs soon after birth, during lactation, and results in dominance of the gut microbiota by *Bifidobacterium*. The second transition occurs during the weaning period, with the introduction of solid foods and continuation of breast milk feeding,^{17–21} and results in the establishment of an adult-type complex microbiome dominated by the phyla *Bacteroidetes* and *Firmicutes*. These alterations continue until three years of age²² and, subsequently, humans acquire stable gut microbiotas maintained in well-balanced host-microbiota symbiotic states called "Enterotypes". Enterotypes are classified into three types that are dominated by *Bacteroides*, *Prevotella*, and other *Firmicutes*.²³ The early establishment of gut microbiota is affected

* Corresponding author.

E-mail address: nakayama@agr.kyushu-u.ac.jp (J. Nakayama).

Peer review under responsibility of Japanese Society of Allergology.

<http://dx.doi.org/10.1016/j.alit.2017.07.010>

1323–8930/Copyright © 2017, Japanese Society of Allergology. Production and hosting by Elsevier B.V. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

by several factors such as delivery mode (cesarean delivery vs. vaginal delivery), breast milk vs. formula feeding, antibiotic usage, and timing of the introduction of solid foods and cessation of milk feeding.²⁴ In addition, intestinal bacterial colonization has profound effects on the development and physiology of the host immune system in early life and affects health and disease in later life. In particular, colonization with intestinal bacteria is necessary for normal immune system development, as indicated by the loss of immune function in germ-free mice.^{25–27} A number of previous studies have shown that intestinal bacteria and their metabolites, including short chain fatty acids (SCFA), play an important role in the proliferation and differentiation of T cells, such as regulatory T (Treg) and helper T (Th) cells, and B cells such as immune globulin (Ig) A- or IgG-secreting B cells.²⁸

Therefore, development of the gut microbiota in infancy occurs during a ‘critical window,’ and a disturbance in this process may cause immune diseases such as food allergies, atopic dermatitis, and asthma.²⁹ To elucidate this association, a large number of studies on microbiotas and microbial ecology, as well as work in animal models and epidemiological surveys, have been conducted. The recent increase in access to next-generation sequencers has allowed researchers to gain deep insight into intestinal ecosystems at the molecular level. On the other hand, hygiene hypothesis has been introduced into gut microbiology to correlate changes in life style with marked increase in childhood allergy in the past half century. This review aims to provide an overview of the recent advances in research on infant gut microbiotas and health and disease, including childhood allergies.

GI tract microbiotas in utero

The GI tract has traditionally been considered sterile until it is colonized by microorganisms residing in the environment at birth. However, recent studies have revealed the presence of microorganisms in amniotic fluid,^{30–32} fetal membranes,³³ umbilical cords, placentas,³⁴ and meconium.^{35–37} Gosalbes *et al.*³⁷ showed that meconium microbiotas could be classified into two types: the first is less diverse and dominated by bacteria in the family Enterobacteriaceae, and the other is more diverse and dominated by bacteria

in the phylum *Firmicutes*, especially lactic acid bacteria. These microbial communities differ from the microbiotas of the vagina, feces, or skin of a pregnant women, but resemble the microbiota of the amniotic fluid, suggesting that microbes in the meconium originate from the uterus of the mother. A fetus's GI tract appeared to be colonized by bacteria through amniotic fluid that was swallowed. Further, the type of microbiota in the meconium was associated with maternal factors such as history of allergies and could have consequences for childhood health.

Experimental work with mice has demonstrated efflux of bacteria from a mother's gut to that of her fetus.³⁸ Pregnant mice were orally inoculated with a genetically labeled *Enterococcus faecium* strain that was previously isolated from the breast milk of a healthy woman. The strain was detected in the amniotic fluid of the inoculated animals. This suggests that bacteria might be transferred from the oral cavity to the uterus, possibly via the blood stream. Oral bacteria are known to reach the uterus via the blood stream, particularly in periodontal disease.³⁹ This route is also supported by the findings of Aagaard *et al.* and others, which has shown that the genetic and taxonomic composition of the placental microbiota closely resemble those of the oral cavity.

Compositional and functional development of the gut microbiota during infancy (Fig. 1)

After birth, the gut microbiota of a newborn is transiently dominated by *Enterobacteriaceae* and *Staphylococcus*.⁴⁰ Thereafter, an infant's gut microbiota is dominated by *Bifidobacterium* and some lactic acid bacteria.¹⁸ *Bifidobacterium*-dominated microbiota, called “Bifidus flora,” is maintained until the introduction of solid food.^{41,42} After weaning, Bifidus flora is outcompeted by adult-type microorganisms, represented mainly by bacteria in the genera *Bacteroides*, *Prevotella*, *Ruminococcus*, *Clostridium*, and *Veillonella*, which colonize an infant's intestines.⁴¹ Eventually, by about three years of age, a typical adult-like gut microbiota is established.²² The functions of the gut microbiota also change greatly before and after the introduction of weaning foods. The functional repertoire of an infant's microbiota changes during the first year of life, as the early microbiota before weaning is enriched in bacteria with genes that

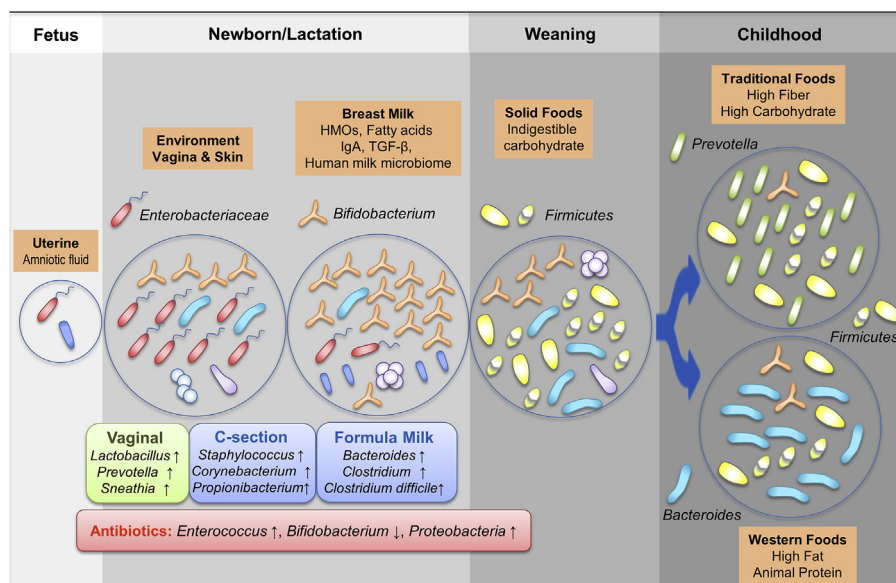


Fig. 1. Fetal-to-childhood gut microbiota colonization and important factors affecting this process. Establishment of the gut microbiota may begin *in utero* and be affected by dynamic shifts in early life. Diversity of the gut microbiota increases with age until it becomes a stable adult microbiota. This process of establishing the gut microbiota is affected by various factors such as delivery mode, methods of milk feeding, the introduction of solid foods, and foods consumed daily in childhood.

facilitate lactate utilization, whereas, after weaning, solid foods promote the growth of bacteria enriched in genes coding to allow the utilization of a larger variety of carbohydrates, vitamin synthesis, and xenobiotic degradation.^{22,41,43}

Effect of the mode of newborn delivery

A number of studies have indicated that the mode of delivery affects the development of the gut microbiota in early life. Interestingly, the gut microbiota of a newborn will closely resemble the microbiota that it encountered during birth. In vaginally delivered infants, gut microbiotas resemble their mothers' vaginal microbiotas, which are dominated by *Lactobacillus*, *Prevotella*, or *Sneathia*, whereas the microbiotas of infants born by caesarean section are most similar to skin microbiota, which is dominated by *Staphylococcus*, *Corynebacterium*, and *Propionibacterium*.³⁶ In addition, some studies have shown that colonization by *Bacteroides* and *Bifidobacterium* is delayed for one month after birth, whereas *Clostridium difficile* was abundant at one month.^{44,45} Underrepresentation of *Bacteroides* was also observed in the microbiotas of infants born by caesarean section for three to four months after birth. In addition, the same studies have shown that infants born by elective caesarean section had particularly low bacterial diversity.⁴⁶ Caesarean birth has been associated with an increased risk for immune disorders such as allergic rhinitis, asthma, and celiac disease,^{47–49} as described in detail below.

Beneficial factors in breast milk

Infant feeding methods, namely breast milk feeding and formula feeding, greatly affect the development of the gut microbiota in early life. Human milk contains proteins, fats, and carbohydrates, as well as immunoglobulins and endocannabinoids. The oligosaccharides in human milk (HMO) such as galactooligosaccharide (GOS), are one of the main components of breast milk. They are only partially digested in the small intestine and mostly reach the colon, where they are fermented, mainly by *Bifidobacterium*, to produce short-chain fatty acids.^{48,50} Sakurama *et al.* reported that *Bifidobacterium* spp. have an enzyme named lacto-*N*-biosidase to facilitate assimilation of GOS.⁵¹ Matsuki *et al.*⁴⁰ reported that, as the number of *Bifidobacterium* increased, the amount of HMOs in feces decreased, and the amount of acetic acid and lactic acid increased in infants at one month of life. Therefore, HMOs have a clear probiotic effect by selectively stimulating the development of a *Bifidobacterium*-rich microbiota.⁴⁰

Recently, milk formula has been improved, notably by the inclusion of some oligosaccharides, which makes it possible for an infant to establish a *Bifidobacterium*-rich microbiota.⁵² However, compared with breastfed infants, formula-fed infants still have distinct features of their microbiotas, such as the overrepresentation of *C. difficile*.⁴⁶ Another study indicated that the microbiotas of formula-fed infants are enriched with anaerobic organisms such as *Bacteroides* and *Clostridium*, whereas those of breastfed infants are more commonly colonized by aerobic organisms.^{53,54} A recent study reported that supplementation with pasteurized donor human milk was partially successful in promoting a microbial community similar to that of breastfed infants and in promoting rapid increases in bacterial diversity.⁵⁵

Breast milk contains numerous factors that modulate and promote the development of the immune system in infancy, including immunoglobulins such as IgA and IgG, antimicrobial compounds such as lysozyme and lactoferrin, immune regulatory cytokines such as TGF- β and interleukin 10 (IL-10), and lymphocytes that express gut homing markers.^{56–58} These substances in breast milk select bacteria that colonize the GI tract. For example, immune

regulatory cytokines such as IL-10 and TGF- β in breast milk facilitate tolerance of the host immune system to intestinal bacteria and promote infant IL-10 production.^{59,60}

Bifidobacterium, whose growth is promoted by HMOs, and especially *Bifidobacterium infantis*, has been reported to directly correlate with the amount of secreted IgA and to have anti-inflammatory effects.^{61,62} Recent studies have shown that breast milk is not sterile. It contains as many as 600 different bacterial species and 10^3 – 10^4 cfu/mL of bacteria cells.⁶³ Genera of bacteria isolated from breast milk include mostly lactic acid bacteria such as *Lactobacillus*, *Leuconostoc*, *Streptococcus*, *Enterococcus*, *Lactococcus*, and *Weissella*, as well as some beneficial *Bifidobacterium* species.^{64–66} In addition, studies using molecular approaches indicate that gram-negative bacteria are also present, including *Serratia*, *Pseudomonas*, and some typical inhabitants of the oral cavity such as *Veillonella*, *Leptotrichia*, and *Prevotella*.⁶⁷ This suggests that some bacteria may be transferred from mother to child via breast milk.

Fatty acids (FA) in breast milk are also associated with host health and development of the immune system. TGF- β levels in breast milk have been reported to correlate positively with polyunsaturated fatty acid content and negatively with saturated FA content.⁶⁸ Some cohort studies have shown that the ratio of polyunsaturated FA is related to an infant's risk of atopic dermatitis.^{68,69} In addition, the microbiotas and FA profiles in milk were different in Europe (Spain and Finland), Africa (South Africa), and Asia (China).⁷⁰ However, there is little understanding of the mechanisms underlying the effects of FA in breast milk on the development of the host immune system, and how they may be related to the establishment of the gut microbiota in infancy.

Perturbation by antibiotics

The use of antibiotic in early life has profound effects on the development of the gut microbiota. The use of antibiotics in infants shifts the composition of the gut microbiota toward a high abundance of Proteobacteria and low abundance of Actinobacteria populations.^{71,72} decreases the overall diversity of the infant's microbiota, and selects for drug-resistant bacteria.^{73,74} According to some epidemiological surveys, the use of antibiotics in early life increases the risk of developing allergic diseases such as asthma, atopic disease, eczema, and type 1 diabetes.⁷⁵

Tanaka *et al.*,⁶⁹ carefully examined the effects of antibiotic exposure in the early postnatal period on the development of the intestinal microbiota. The fecal microbiotas of newborns orally administered a broad-spectrum antibiotic for the first four days of life were analyzed for two months. In the first week of the infants' lives, the antibiotic-administered subjects showed less diversity in their fecal bacterial communities, with the attenuation of some bacterial groups, especially *Bifidobacterium*, as well as unusual colonization with *Enterococcus*. At one month of age, overgrowth of Enterobacteriaceae and *Enterococcus* was observed in infants in the antibiotic-treated group. Interestingly, caesarean-delivered subjects whose mothers were intravenously administered a closely related antibiotic in the same period showed similar, although weaker, associations with microbiota development. These results indicate that antibiotic exposure at the beginning of life greatly affects the development of neonatal intestinal microbiotas.

Introduction of solid food

A large shift in the gut microbial community from *Bifidobacterium*-dominant to *Bacteroidetes*- and *Firmicutes*-dominant accompanies the introduction of solid foods, including indigestible carbohydrates, into an infant's diet, although the specific taxa present before and after this point varies.^{41,42,76} At approximately

three years of age, bacterial composition and diversity is most like those of adults.²² These microbiotas remain fairly stable throughout adulthood in the absence of perturbations such as long-term dietary changes, disease-associated dysbiosis, or the use of antibiotics.

Recent metagenomics studies have offered in-depth insights into the functions of infant gut microbiotas. The infant gut rapidly acquires a functional gene pool dominated by carbohydrate metabolism genes, which is broadly similar to that of an adult.²² However, the functional repertoires of the infant microbiotas shift dramatically during the first year of life, as the earliest microbiotas are enriched in bacteria with genes that facilitate lactate utilization, whereas solid foods enrich microbiotas with bacteria with genes that code for the utilization of a larger variety of carbohydrates, vitamin biosynthesis, and xenobiotic degradation.^{19,43}

Childhood microbiotas

The gut microbiomes of children, compared with those of infants, show less variability among individuals and are more stable. Gut microbiotas in childhood are affected by geography and food culture. Nakayama *et al.* taxonomically assigned the gut microbiotas of 303 children from five Asian countries, namely Japan, China, Taiwan, Indonesia, and Thailand, and found that they clustered into *Bacteroides-Bifidobacterium*-dominate microbiotas (BB-type) and *Prevotella*-dominated microbiotas (P-type).⁷⁷ The BB-type was most common in Eastern Asian, whereas the P-type was predominant in central Asia and South East Asia, with Indonesia having the highest abundance of P-type microbiotas. The establishment of two distinct intestinal microbial communities may be largely influenced by foods consumed daily, as shown by the P-type that is established in vegetarians.⁷⁸

The two types of microbiotas have been reported in many other cohort studies. Yatsunenko *et al.* characterized bacterial species in fecal samples from 531 individuals comprising healthy children and adults from the Amazonas of Venezuela, rural Malawi, and metropolitan USA.²² The gut microbiotas of people from the USA were dominated by *Bacteroides*, whereas those of rural Venezuela and Malawi people were dominated by *Prevotella*. Lin *et al.* compared microbiotas of children in the USA and Bangladesh and found that Bangladeshi children had high bacterial diversity, with communities dominated by *Prevotella*, whereas US children had low bacterial diversity, with communities dominated by *Bacteroides* and *Firmicutes*.⁷⁹ These results suggested that gut microbiotas of children in advanced countries are dominated by *Bacteroides* and *Firmicutes*, whereas those of developing countries are dominated by *Prevotella*. Further, these two microbiota-types are often observed within the same country, but are associated

with level of development in an area. Nakayama *et al.* recently found that P- and BB-type microbiotas were represented in children living in rural and urban sites, respectively, on Leyte island in Philippines, and that a type shift was associated with modernization of consumed foods, and notably a change to high-fat diets.⁸⁰ The two sites sampled, namely Baybay and Ormoc, were only 50 km apart, and the children studied were mostly Filipino, suggesting that diet had a greater influence on gut microbial communities than host genetics. Indeed, the functional repertoires of the microbiotas of children on Leyte reflected dietary habit, and the microbiotas were enriched in bacteria with genes encoding bile acids that aid in lipid absorption or oligosaccharide-degrading enzymes involved in plant digestion in BB-type and P-type children, respectively.

It is notable that gut microbiotas differ between advanced and developing countries and between urban and rural areas, and that the occurrence of autoimmune diseases such as allergic diseases or IBD has coincided with industrialization in the past half century. Many papers have suggested the association of gut microbiota alterations with the development of allergies, which is expressed in the gut microbiota-mediated hygiene hypothesis.

Interactions between gut microbiotas and host immunity (Fig. 2)

Development of gut microbiotas between infancy and weaning is associated with immune system development.²⁴ In particular, bacterial colonization is indispensable for the normal development of immunity, as indicated by the loss of immune function in germ-free mice.^{25–27,81} Furthermore, bidirectional communications have been proposed, in which intestinal bacteria affect not only the development of the immune system but also the host immune system influences the development of the gut microbiota.⁸²

Immune factors in infancy, such as the presence of maternal antibodies and intestinal gut microbiota-primed B cell trafficking, play important roles in the selection of intestinal bacteria.^{19,28,83} Through the placenta and breast milk, maternal antibodies such as IgG and IgA are introduced into infants.^{56–58} The presence of IgAs specific for intestinal bacteria are associated with differences in bacteria habitats in GI tracts. Bunker *et al.* showed that intestinal bacteria in the small intestine are bound to bacteria-specific IgA, whereas, in the large intestine, most bacteria are not bound to IgA.⁸⁴ This result suggests that bacteria bound by IgA effectively colonize the small intestine. Furthermore, segmented filamentous bacteria (SFB) over colonize the GI tracts of IgA-knockout mice and strongly induce immunity.^{85,86} SFB play a role in shaping immune functions, such as promoting IgA secretion^{86,87} and promoting the development of intestinal cell lymphocytes and Th17 cells.⁸⁸ Based

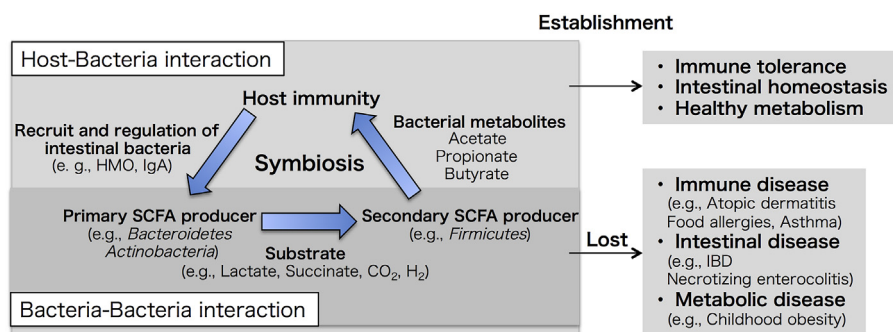


Fig. 2. Infant's gut microbiota and the host immune system interaction. The resulting gut microbial community and metabolites contributes to the development of the host immune system, intestinal homeostasis and host metabolism.

on these functions of IgA molecules, maternal IgA in breast milk functions similarly; thus, IgA may be involved in the selection of bacteria that colonize GI tracts in infancy.

Bacterial colonization of the gut also impacts differentiation of naïve T cells into Forkhead box P3 (Foxp3)⁺ Treg cells or various types of Th cells such as Th1, Th2, and Th17^{82,89}. Treg cells suppress the differentiation of naïve T cells into Th cells^{90,91} and have various anti-inflammatory effects, including suppression of the inflammatory activities of mast cells, basophils, and eosinophils, suppression of IgE, and induction of IgG4.⁹² A number of intestinal bacteria, including *Lactobacillus*, *Bifidobacterium*, *Bacteroides*, *Clostridium*, and *Streptococcus*,⁹³ as well as bacterial metabolites such as butyric acid and propionic acid, have been shown to induce Treg cells in various mouse models or cell culture.⁹⁴ Treg cells are generated in the thymus (tTreg) and periphery (pTreg), and pTregs, but not tTregs, control mucosal Th2 inflammation in the gut and lungs.⁹⁵ Recent studies have shown that pTregs are promoted by intestinal bacteria and a symbiosis factor (polysaccharide A) produced by *Bacteroides fragilis*.^{96,97} On the other hand, each type of Th cell plays a distinct and key role in shaping and amplifying the immune response by producing cytokines that can suppress other types of Th cells. Th17 cells, which are abundant in the mucosa, secrete cytokines, including IL-17, IL-17F, and IL-22. These cytokines improve the barrier function of the GI tract and protect against pathogen and fungi. Th17 cells are promoted by bacterial flagella, unmethylated DNA, and adenosine triphosphate (ATP).²⁷ Mutual regulation of Th1 and Th2 cells is considered a critical factor for immune homeostasis. Indeed, excessive Th1 or Th2 activation results in chronic inflammation and autoimmune or allergic disease.

Gut microbiota aberrations associated with allergy development

Colonization of intestines with bacteria and the development of a gut microbiota in infancy are closely related to the development of the immune system.^{15,24,29,98,99} In fact, altered development of gut microbiotas is suspected to be associated with the onset of autoimmune or allergic diseases.

Numerous cohort studies have shown that alterations in the gut microbiota during infancy and early childhood are associated with allergic diseases (Table 1). Atopic eczema/dermatitis reflects inflammation of the skin and is the result of a process called atopic march, which constitutes a strong risk factor for the development of other allergic disease.⁹⁸ Some cohort studies have shown that infants with atopic dermatitis have a low level of bacterial diversity in early infancy, as well as a low abundance of *Bifidobacterium* and *Bacteroides* and high abundance of *Enterobacteriaceae*.^{100–104} Fujimura *et al.* reported that infants with high relative risk of atopic dermatitis and asthma showed lower relative abundances of certain bacteria such as *Bifidobacterium*, *Akkermansia*, and *Faecalibacterium*, high relative abundances of particular fungi such as *Candida* and *Rhodotorula*, and distinct fecal metabolites enriched with pro-inflammatory metabolites, 12, 13-DiHOME.¹⁰⁵

Food allergies are a disease with symptoms such as inflammation of the skin and GI tract resulting from specific food intake.¹⁰⁶ Symptoms, in most cases, appear when solid foods are introduced in association with weaning. Food allergies in infancy are thought to impair oral tolerance. Establishment of oral tolerance is a crucial event mediated by bacterial colonization in infancy, and it is required for normal function of the digestive tract after weaning.

Table 1
Prospective cohort studies that characterized gut microbiota associated with allergies.

Surveyed country	Allergic disease	Age	No.	Features of gut microbial community or metabolites†	Ref. ‡
Sweden Estonia	Atopic dermatitis	–y2	44	<i>Enterococcus</i> ↓ (m1), <i>Bifidobacterium</i> ↓ (y1), <i>Clostridium</i> ↑ (m3), <i>Bacteroides</i> ↓ (y1), <i>Staphylococcus aureus</i> ↑ (m6)	¹⁰³ (2001)
Finland	Atopic dermatitis	Lactation	37	Bacterial no. ↓ (m5, m6), <i>Bacteroides</i> ↑ (m5, m6), Gram positive aerobic bacteria ↓ (m5, m6), <i>Bifidobacterium</i> ↓ (m5, m6)	¹¹⁷ (2001)
Finland	Atopic dermatitis	–y1	29	<i>Clostridium</i> ↑ (w3), <i>Bifidobacterium</i> ↓ (w3)	¹⁰¹ (2001)
Finland	Atopic dermatitis	m2–m7	10	<i>Bifidobacterium adolescentis</i> ↑, <i>Bifidobacterium bifidum</i> ↓	¹¹⁸ (2001)
Japan	Atopic dermatitis	–y2	12	<i>Bacteroides to Bifidobacterium</i> ↑ (y2)	¹¹⁹ (2005)
Estonia	Food allergies	y1, y4	139 (y1)	Acetate ↑ (y4) Butyrate ↓ (y1, y4)	¹¹⁶ (2013)
Sweden	Atopic dermatitis	y1, y4	53 (y4)	Valerate ↓ (y1, y4)	
Japan	Food allergies	w1, m1, m2	22	<i>Acinetobacter</i> ↓ (m1), <i>Clostridium</i> ↓ (m1) <i>Bacteroides</i> ↑ (m1), <i>Propionibacterium</i> ↑ (m2) <i>Klebsiella</i> ↑ (m2)	¹²⁰ (2009)
Sweden	Atopic dermatitis	w1, m1, y1	40	Bacterial diversity ↓ (m1) <i>Bacteroides</i> diversity ↓ (m1) Proteobacteria diversity ↓ (m12)	¹⁰⁴ (2012)
China	Food allergies	m2–m11	79	<i>Bacteroides</i> ↓, <i>Clostridium XVIII</i> ↓, <i>Clostridium sensu stricto</i> ↑, <i>Anaerobacter</i> ↑	¹⁰⁷ (2014)
Canada	Food sensitization	m3, y1	166	Bacterial Diversity ↓ (m3), <i>Bacteroides</i> ↓ (y1) <i>Enterobacteriaceae</i> ↑ (m3)	¹⁰⁸ (2015)
Canada	Asthma Wheeze	m3, y1	319	<i>Veillonella</i> ↓ (m3, y1), <i>Lachnospira</i> ↓ (m3, y1) <i>Rohia</i> ↓ (m3), <i>Faecalibacterium</i> ↓ (m3)	¹²¹ (2015)
USA	Milk allergy resolve	m3–m16	226	<i>Clostridiaceae</i> ↑ (m3–m6) Fatty acid associated genes ↑ (m3–m6) <i>Bacteroidales</i> ↓ (m3–m6)	¹¹⁰ (2016)
USA	Atopic dermatitis	m1–m11	298	<i>Bifidobacterium</i> ↓, <i>Akkermansia</i> ↓, <i>Faecalibacterium</i> ↓, <i>Candida</i> ↑, <i>Rhodotorula</i> ↑, 12,13-DiHOME ↑	¹⁰⁵ (2016)
Taiwan	Food sensitization	Weaning	45	<i>Bacteroidetes</i> ↓, <i>Veillonella</i> ↓ <i>Bifidobacterium</i> ↑, <i>Firmicutes</i> ↑	¹²² (2016)
Australia	Atopic dermatitis Eczema	w1, m1, m3	117	<i>Bifidobacterium breve</i> ↓ (w1, m1, m3) <i>Bifidobacterium catenulatum</i> ↑ (m1, m3)	¹²³ (2016)

† y, year of age; m, month of age; w, weak of age; ↑, high abundance; ↓, low abundance.

‡ The number in parentheses indicates the year of publication.

Ling *et al.* showed that infants with food allergies before one year of age have a specific gut microbiota showing lower relative abundances of *Bacteroides* and *Clostridium* XVIII and higher relative abundance of *Clostridium sensu stricto* and *Anaerobacter*.¹⁰⁷ Azad *et al.* studied infants with food sensitivity at one year of life and found that they had low-diversity and *Enterobacteriaceae*-dominated microbiota at three months of age and showed higher abundances of *Bacteroides* at one year of age, relative to children without food sensitivities.¹⁰⁸

The authors of this review found that the gut microbiotas of infants with food allergies during the first two years of life showed lower relative abundances of some lactic acid bacteria and *Veillonella* in the lactation period, and had low-diversity and *Enterobacteriaceae*-dominant microbiotas, with colonization by *Clostridium paraputrificum* and *Clostridium tertium*.¹⁰⁹ On the other hand, Bunyanuvanch *et al.* conducted a cohort study examining milk allergies and found that children who resolved milk allergies by the age of eight years had microbiotas with higher abundances of bacteria in the phylum *Firmicutes* and family *Clostridiaceae* during the lactation period, whereas the microbiotas of children with persistent milk allergies showed higher abundances of *Bacteroides*.¹¹⁰

Gut microbiota-mediated hygiene hypothesis

The number of people with allergies rose drastically in the past half century after world industrialization. This phenomenon is often explained by the “hygiene hypothesis,” which states that less exposure to parasites and microbes leads to an overreactive immune system. An imbalance between type 1 and type 2 Th cells can indeed interpret allergy crisis in advanced countries with modern hygiene environment that biases humoral immunity in human body. With the eradication of certain pathogens, less exposure to environmental and commensal microorganisms may contribute to the effects hypothesized. Indeed, the prevalence of allergic diseases was found to be significantly higher in urban children than in rural children,¹¹¹ although the environment where the rural children were living also had a much higher level of bacterial endotoxin.¹¹² Interestingly, *Acinetobacter lwoffii* F78, isolated from a farming environment, showed a potent allergy-protection effect by inducing Th1 polarization in the respiratory system.¹¹³

The hygiene hypothesis has been extended to intestinal microbiology. Björkstén *et al.*¹⁰³ examined differences in gut microbial composition between allergic and non-allergic children, as well as between children living in a developed country, Sweden, and a developing country, Estonia. Following this study, a number of epidemiological studies have found differences in gut microbial composition between allergic and non-allergic children, even though the differences were not always consistent. The discrepancies may result from differences in races, genetics, diets, living environments, or other factors, in addition to study methodologies, including sampling ages or analytical methods used to investigate microbial composition. Our group has also addressed differences in gut microbiotas between one-month-old Japanese infants who later did and did not develop allergic diseases.¹¹⁴ Indeed, allergic infants were more commonly colonized by bacteria in the genus *Bacteroides* and less often colonized by bacteria in the genera *Acinetobacter* and *Clostridium*. It is interesting to find that *Acinetobacter* was a less frequent colonizer of allergic infants in Japan.

Recently, Vatanen *et al.*¹¹⁵ followed gut microbiota development from birth until age three years in 222 infants in Northern Europe. Early-onset autoimmune diseases are common in Finland and Estonia, but are less prevalent in Russia. Interestingly, they found that *Bacteroides* species were not abundant in Russians, but dominate in Finnish and Estonian infants. This coincides with our

previous finding that a high abundance of *Bacteroides* was associated with the development of allergies in Japanese infants. *Bacteroides* LPS is known to be structurally distinct from that of *Escherichia coli*, and it inhibits innate immune signaling and endotoxin tolerance, which is normally induced by commensal members of gram-negative bacteria such as *E. coli*. Early colonization by such immune-silencing microorganisms may have precluded aspects of immune education in allergic children in Finland and Estonia.

Childhood food allergies have emerged in a second wave of the allergy epidemic, following a first wave of asthma, atopic dermatitis, and allergic rhinitis that appeared in association with industrialization in the past half century.¹¹⁶ However, the cause of this second epidemic is yet to be identified. Recently, omics technology, especially metagenomics and metabolomics, has progressed dramatically, facilitating examinations of the structural and functional aspects of gut microbial communities, and advanced animal experiment technologies are now available that allow us to gain insight into the cellular and molecular mechanisms underlying the associations between gut microbiotas and childhood allergies.

Concluding remarks

Stable microbial communities are established through dynamic changes in infancy and are affected by various environmental factors such as the mode of delivery and methods of milk feeding. Simultaneously, the immune system matures through bidirectional interactions, with the gut microbiota promoting the development of the immune system but also the host immune system shaping the development of the gut microbiota. Alterations in the development of gut microbiotas during infancy can have a variety of negative effects on immune health and the onset of allergies. Although our knowledge of these effects is far from complete, many studies are now allowing us to determine the causes and effects described in the gut microbiota-mediated hygiene hypothesis. These insights may help us develop a society with less allergy diseases than in the pre-industrialized era.

Acknowledgements

This study was supported in part by Grants-in-Aid for Scientific Research (B) No. 25304006 and No. 17H04620 from the Japan Society for the Promotion of Science (JSPS) (to Jiro Nakayama).

Conflict of interest

The authors have no conflict of interest to declare.

References

1. Round JL, Mazmanian SK. The gut microbiome shapes intestinal immune responses during health and disease. *Nat Rev Immunol* 2009;9:313–23.
2. Nicholson JK, Holmes E, Kinross J, Burcelin R, Gibson G, Jia W, et al. Host-gut microbiota metabolic interactions. *Science* 2012;308:1262–8.
3. Bäckhed F, Ley RE, Sonnenburg JL, Peterson DA, Gordon JL. Host-bacterial mutualism in the human intestine. *Science* 2005;307:1915–20.
4. Turnbaugh PJ, Ridaura VK, Faith JJ, Rey FE, Knight R, Gordon JL. The effect of diet on the human gut microbiome: a metagenomic analysis in humanized gnotobiotic mice. *Sci Trans Med* 2009;1:6ra14.
5. Claus SP, Guillou H, Ellero-Simatos S. The gut microbiota: a major player in the toxicity of environmental pollutants? *NPJ Biofilms Microbiomes* 2016;2:16003.
6. Sakamoto I, Igarashi M, Kimura K, Takagi, Miwa T, Koga Y. Suppressive effect of *Lactobacillus gasseri* OLL 2716 (LG21) on *Helicobacter pylori* infection in humans. *J Antimicrob Chemother* 2001;47:709–10.
7. Matsui H, Takahashi T, Øverby A, Murayama SY, Yoshida H, Yamamoto Y, et al. Mouse models for assessing the protective efficacy of *Lactobacillus gasseri* SBT2055 against *Helicobacter suis* infection associated with the development of gastric mucosa-associated lymphoid tissue lymphoma. *Helicobacter* 2015;20:291–8.
8. Hooper LV, Macpherson AJ. Immune adaptations that maintain homeostasis with the intestinal microbiota. *Nat Rev Immunol* 2010;10:159–69.

9. Quince C, Ijaz UZ, Loman N, Eren AM, Saulnier D, Russell J, et al. Extensive modulation of the fecal metagenome in children with crohn's disease during exclusive enteral nutrition. *Am J Gastroenterol* 2015;**110**:1718–29.
10. Codling C, O'Mahony L, Shanahan F, Quigley EMM, Marchesi JR. A molecular analysis of fecal and mucosal bacterial communities in irritable bowel syndrome. *Dig Dis Sci* 2010;**55**:392–7.
11. Kadooka Y, Sato M, Imaizumi K, Ogawa A, Ikuyama K, Akai Y, et al. Regulation of abdominal adiposity by probiotics (*Lactobacillus gasseri* SBT2055) in adults with obese tendencies in a randomized controlled trial. *Eur J Clin Nutr* 2010;**64**:636–43.
12. Kang JH, Yun S II, Park MH, Park JH, Jeong SY, Park HO. Anti-obesity effect of *Lactobacillus gasseri* BNR17 in high-sucrose diet-induced obese mice. *PLoS One* 2013;**8**:1–8.
13. Miyoshi M, Ogawa A, Higurashi S, Kadooka Y. Anti-obesity effect of *Lactobacillus gasseri* SBT2055 accompanied by inhibition of pro-inflammatory gene expression in the visceral adipose tissue in diet-induced obese mice. *Eur J Nutr* 2014;**53**:599–606.
14. Karimi G, Sabran MR, Jamaluddin R, Parvaneh K, Mohtarrudin N, Ahmad Z, et al. The anti-obesity effects of *Lactobacillus casei* strain Shirota versus Orlistat on high fat diet-induced obese rats. *Food Nutr Res* 2015;**59**:1–9.
15. Lynch SV, Boushey HA. The Microbiome and development of allergic disease. *Curr Opin Allergy Clin Immunol* 2016;**16**:165–71.
16. Wang Y, Kasper LH. The role of microbiome in central nervous system disorders. *Brain Behav Immun* 2014;**38**:1–12.
17. Bergström A, Skov TH, Bahl MI, Roager HM, Christensen LB, Ejlerskov KT, et al. Establishment of intestinal microbiota during early life: a longitudinal, explorative study of a large cohort of Danish infants. *Appl Environ Microbiol* 2014;**80**:2889–900.
18. Mitsuoka T. Development of functional foods. *Biosci Microbiota Food Health* 2014;**33**:117–28.
19. Bäckhed F, Roswall J, Peng Y, Feng Q, Jia H, Kovatcheva-Datchary P, et al. Dynamics and stabilization of the human gut microbiome during the first year of life. *Cell Host Microbe* 2015;**17**:690–703.
20. Dogra S, Sakwinska O, Soh SE, Ngom-Bru C, Brück WM, Berger B, et al. Rate of establishing the gut microbiota in infancy has consequences for future health. *Gut Microbes* 2015;**6**:321–5.
21. Dogra S, Sakwinska O, Soh S-E, Ngom-Bru C, Brück WM, Berger B, et al. Dynamics of Infant gut microbiota are influenced by delivery mode and gestational duration and are associated with subsequent. *Gut Microbes* 2015;**6**:1–9.
22. Yatsunencko T, Rey FE, Manary MJ, Trehan I, Dominguez-Bello MG, Contreras M, et al. Human gut microbiome viewed across age and geography. *Nature* 2012;**486**:222–7.
23. Arumugam M, Raes J, Pelletier E, Le Paslier D, Yamada T, Mende DR, et al. Enterotypes of the human gut microbiome. *Nature* 2011;**473**:174–80.
24. Francino M. Early development of the gut microbiota and immune health. *Pathogens* 2014;**3**:769–90.
25. Purchiaroni F, Tortora A, Gabrielli M, Bertucci F, Gigante G, Ianiro G, et al. The role of intestinal microbiota and the immune system. *Eur Rev Med Pharmacol Sci* 2013;**17**:323–33.
26. Macpherson AJ, Harris NL. Interactions between commensal intestinal bacteria and the immune system. *Nat Rev Immunol* 2004;**4**:478–85.
27. Atarashi K, Nishimura J, Shima T, Umesaki Y, Yamamoto M, Onoue M, et al. ATP drives lamina propria T(H)17 cell differentiation. *Nature* 2008;**455**:808–12.
28. Nguyen QN, Himes JE, Martinez DR, Permar SR. The impact of the gut microbiota on humoral immunity to pathogens and vaccination in early infancy. *PLoS Pathog* 2016;**12**:e1005997.
29. Stiemsma LT, Turvey SE. Asthma and the microbiome: defining the critical window in early life. *Allergy Asthma Clin Immunol* 2017;**13**:1–9.
30. DiGiulio DB, Romero R, Amogan HP, Kusanovic JP, Bik EM, Gotsch F, et al. Microbial prevalence, diversity and abundance in amniotic fluid during preterm labor: a molecular and culture-based investigation. *PLoS One* 2008;**3**:1–10.
31. DiGiulio DB, Romero R, Kusanovic JP, Gómez R, Kim CJ, Seok KS, et al. Prevalence and diversity of microbes in the amniotic fluid, the fetal inflammatory response, and pregnancy outcome in women with preterm pre-labor rupture of membranes. *Am J Reprod Immunol* 2010;**64**:38–57.
32. Romero R, Miranda J, Kusanovic JP, Chaiworapongsa T, Chaemsaitong P, Martinez A, et al. Clinical chorioamnionitis at term I: microbiology of the amniotic cavity using cultivation and molecular techniques. *J Perinat Med* 2015;**43**:19–36.
33. Jiménez E, Fernández L, Marín ML, Martín R, Odriozola JM, Nueno-Palop C, et al. Isolation of commensal bacteria from umbilical cord blood of healthy neonates born by cesarean section. *Curr Microbiol* 2005;**51**:270–4.
34. Aagaard K, Ma J, Antony KM, Ganu R, Petrosino J, Versalovic J. The placenta harbors a unique microbiome. *Sci Trans Med* 2014;**6**:237ra65.
35. Moles L, Gómez M, Heilig H, Bustos G, Fuentes S, de Vos W, et al. Bacterial diversity in meconium of preterm neonates and evolution of their fecal microbiota during the first month of life. *PLoS One* 2013;**8**:e66986.
36. Dominguez-Bello MG, Costello EK, Contreras M, Magris M, Hidalgo G, Fierer N, 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**:11971–5.
37. Gosalbes MJ, Llop S, Vallès Y, Moya A, Ballester F, Francino MP. Meconium microbiota types dominated by lactic acid or enteric bacteria are differentially associated with maternal eczema and respiratory problems in infants. *Clin Exp Allergy* 2013;**43**:198–211.
38. Jiménez E, Marín ML, Martín R, Odriozola JM, Olivares M, Xaus J, et al. Is meconium from healthy newborns actually sterile? *Res Microbiol* 2008;**159**:187–93.
39. Kormman KS, Loesche WJ. The subgingival microbial flora during pregnancy. *J Periodontol* 1980;**15**:111–22.
40. Matsuki T, Yahagi K, Mori H, Matsumoto H, Hara T, Tajima S, et al. A key genetic factor for fucosylactose utilization affects infant gut microbiota development. *Nat Commun* 2016;**7**:11939.
41. Vallès Y, Artacho A, Pascual-García A, Ferrús ML, Gosalbes MJ, Abellán JJ, et al. Microbial succession in the gut: directional trends of taxonomic and functional change in a birth cohort of Spanish infants. *PLoS Genet* 2014;**10**:e1004406.
42. Palmer C, Bik EM, DiGiulio DB, Relman DA, Brown PO. Development of the human infant intestinal microbiota. *PLoS Biol* 2007;**5**:1556–73.
43. Vaishampayan PA, Kuehl JV, Froula JL, Morgan JL, Ochman H, Francino MP. Comparative metagenomics and population dynamics of the gut microbiota in mother and infant. *Genome Biol Evol* 2010;**2**:53–66.
44. Penders J, Thijs C, Vink C, Stelma FF, Snijders B, Kummeling I, et al. Factors influencing the composition of the intestinal microbiota in early infancy. *Pediatrics* 2006;**118**:511–21.
45. Biasucci G, Benenati B, Morelli L, Bessi E, Boehm G. Cesarean delivery may affect the early biodiversity of intestinal bacteria. *J Nutr* 2008;**138**:17965–1800.
46. Azad MB, Konya T, Maughan H, Guttman DS, Field CJ, Chari RS, et al. Gut microbiota of healthy Canadian infants: profiles by mode of delivery and infant diet at 4 months. *CMAJ* 2013;**185**:385–94.
47. Decker E, Hornef M, Stockinger S. Cesarean delivery is associated with celiac disease but not inflammatory bowel disease in children. *Gut Microbes* 2011;**2**:91–8.
48. Marcobal A, Barboza M, Froehlich JW, Block DE, German JB, Lebrilla CB, et al. Consumption of human milk oligosaccharides by gut-related microbes. *J Agric Food Chem* 2010;**58**:5334–40.
49. Renz-Polster H, David MR, Buist AS, Vollmer WM, O'Connor EA, Frazier EA, et al. Caesarean section delivery and the risk of allergic disorders in childhood. *Clin Exp Allergy* 2005;**35**:1466–72.
50. Huérou-Luron IL, Blat S, Boudry G. Breast- v. formula-feeding: impacts on the digestive tract and immediate and long-term health effects. *Nutr Res Rev* 2010;**23**:23–36.
51. Sakurama H, Kiyohara M, Wada J, Honda Y, Yamaguchi M, Fukiya S, et al. Lacto- N -biosidase encoded by a novel gene of *Bifidobacterium longum* sub-species *longum* shows unique substrate specificity and requires a designated chaperone for its active expression. *J Biol Chem* 2013;**288**:25194–206.
52. Veeraman-Wauters G, Staelens S, Van de Broek H, Plaskie K, Wesling F, Roger LC, et al. Physiological and bifidogenic effects of prebiotic supplements in infant formulae. *J Pediatr Gastroenterol Nutr* 2011;**52**:763–71.
53. Stark PL, Lee A. *Clostridia* isolated from the feces of infants during the first year of life. *J Pediatr* 1982;**100**:362–5.
54. Marcobal A, Barboza M, Sonnenburg ED, Pudlo N, Martens EC, Desai P, et al. *Bacteroides* in the infant gut consume milk oligosaccharides via mucus-utilization pathways. *Cell Host Microbe* 2011;**10**:507–14.
55. Gregory KE, Samuel BS, Houghteling P, Shan G, Ausubel FM, Sadreyev RI, et al. Influence of maternal breast milk ingestion on acquisition of the intestinal microbiome in preterm infants. *Microbiome* 2016;**4**:68.
56. Iyengar SR, Walker WA. Immune factors in breast milk and the development of atopic disease. *J Pediatr Gastroenterol Nutr* 2012;**55**:641–7.
57. Field CJ. The immunological components of human milk and their effect on immune development in infants. *J Nutr* 2005;**135**:1–4.
58. Palmeira P, Carneiro-Sampaio M. Immunology of breast milk. *Rev Assoc Med Bras* 2016;**62**:584–93.
59. Levast B, Li Z, Madrenas J. The role of IL-10 in microbiome-associated immune modulation and disease tolerance. *Cytokine* 2015;**75**:291–301.
60. Brandtzaeg P. Mucosal immunity: integration between mother and the breast-fed infant. *Vaccine* 2003;**21**:3382–8.
61. Sjögren YM, Tomicic S, Lundberg A, Böttcher MF, Björkstén B, Sverremark-Ekström E, et al. Influence of early gut microbiota on the maturation of childhood mucosal and systemic immune responses: gut microbiota and immune responses. *Clin Exp Allergy* 2009;**39**:1842–51.
62. Chichlowski M, Lartigue GD, German JB, Raybould HE, Mills DA. Bifidobacteria isolated from infants and cultured on human milk oligosaccharides affect intestinal epithelial function. *J Pediatr Gastroenterol Nutr* 2012;**55**:321–7.
63. Jeurink PV, van Bergenhenegouwen J, Jiménez E, Knippels LMJ, Fernández L, Garssen J, et al. Human milk: a source of more life than we imagine. *Benef Microbes* 2013;**4**:17–30.
64. Soto A, Martín V, Jiménez E, Mader I, Rodríguez JM, Fernández L. Lactobacilli and bifidobacteria in human breast milk. *J Pediatr Gastroenterol Nutr* 2014;**59**:78–88.
65. Heikkilä MP, Saris PEJ. Inhibition of *Staphylococcus aureus* by the commensal bacteria of human milk. *J Appl Microbiol* 2003;**95**:471–8.
66. Jost T, Lacroix C, Braegger C, Chassard C. Assessment of bacterial diversity in breast milk using culture-dependent and culture-independent approaches. *Br J Nutr* 2013;**110**:1253–62.

67. Cabrera-Rubio R, Collado MC, Laitinen K, Salminen S, Isolauri E, Mira A. The human milk microbiome changes over lactation and is shaped by maternal weight and mode of delivery. *Am J Clin Nutr* 2012;**96**:544–51.
68. Laiho K, Lampi AM, Hämäläinen M, Moilanen E, Piironen V, Arvola T, et al. Breast milk fatty acids, eicosanoids, and cytokines in mothers with and without allergic disease. *Pediatr Res* 2003;**53**:642–7.
69. Hanson LA, Korotkova M, Teleme E. Breast-feeding, infant formulas, and the immune system. *Ann Allergy Asthma Immunol* 2003;**90**:59–63.
70. Kumar H, du Toit E, Kulkarni A, Aakko J, Linderborg KM, Zhang Y, et al. Distinct patterns in human milk microbiota and fatty acid profiles across specific geographic locations. *Front Microbiol* 2016;**7**:1519.
71. Tanaka S, Kobayashi T, Songjinda P, Tateyama A, Tsubouchi M, Kiyohara C, et al. Influence of antibiotic exposure in the early postnatal period on the development of intestinal microbiota. *FEMS Immunol Med Microbiol* 2009;**56**:80–7.
72. Fouhy F, Guinane CM, Hussey S, Wall R, Ryan CA, Dempsey EM, et al. High-throughput sequencing reveals the incomplete, short-term recovery of infant gut microbiota following parenteral antibiotic treatment with ampicillin and gentamicin. *Antimicrob Agents Chemother* 2012;**56**:5811–20.
73. Greenwood C, Morrow AL, Lagomarcino AJ, Altaye M, Taft DH, Yu Z, et al. Early empiric antibiotic use in preterm infants is associated with lower bacterial diversity and higher relative abundance of enterobacter. *J Pediatr* 2014;**165**:23–9.
74. Moore AM, Ahmadi S, Patel S, Gibson MK, Wang B, Ndao MI, et al. Gut resistome development in healthy twin pairs in the first year of life. *Microbiome* 2015;**3**:27.
75. Langdon A, Crook N, Dantas G. The effects of antibiotics on the microbiome throughout development and alternative approaches for therapeutic modulation. *Genome Med* 2016;**8**:39.
76. Fallani M, Amarri S, Uusijarvi A, Adam R, Khanna S, Aguilera M, et al. Determinants of the human infant intestinal microbiota after the introduction of first complementary foods in infant samples from five European centres. *Microbiology* 2011;**157**:1385–92.
77. Nakayama J, Watanabe K, Jiang J, Matsuda K, Chao S-H, Haryono P, et al. Diversity in gut bacterial community of school-age children in Asia. *Sci Rep* 2015;**5**:8397.
78. Ruengsomwong S, La-Ongkham O, Jiang J, Wannissorn B, Nakayama J, Nitisinprasert S. Microbial community of healthy Thai vegetarians and non-vegetarians, their core gut microbiota, and pathogen risk. *J Microbiol Biotechnol* 2016;**26**:1723–35.
79. Lin A, Bik EM, Costello EK, Dethlefsen L, Haque R, Relman DA, et al. Distinct distal gut microbiome diversity and composition in healthy children from Bangladesh and the United States. *PLoS One* 2013;**8**:e53838.
80. Nakayama J, Yamamoto A, Palermo-Conde LA, Higashi K, Sonomoto K, Tan J, et al. Impact of westernized diet on gut microbiota in children on Leyte Island. *Front Microbiol* 2017;**8**:1–18.
81. Sudo N, Sawamura S, Tanaka K, Aiba Y, Kubo C, Koga Y. The requirement of intestinal bacterial flora for the development of an IgE production system fully susceptible to oral tolerance induction. *J Immunol* 1997;**159**:1739–45.
82. Belkaid Y, Hand TW. Role of the microbiota in immunity and inflammation. *Cell* 2014;**157**:121–41.
83. Clemente JC, Ursell LK, Parfrey LW, Knight R. The impact of the gut microbiota on human health: an integrative view. *Cell* 2012;**148**:1258–70.
84. Bunker JJ, Flynn TM, Koval JC, Shaw DG, Meisel M, McDonald BD, et al. Innate and adaptive humoral responses coat distinct commensal bacteria with immunoglobulin A. *Immunity* 2015;**43**:541–53.
85. Fagarasan S, Muramatsu M, Suzuki K, Nagaoka H, Hiai H, Honjo T. Critical roles of activation-induced cytidine deaminase in the homeostasis of gut flora. *Science* 2002;**298**:1424–7.
86. Suzuki K, Meek B, Doi Y, Muramatsu M, Chiba T, Honjo T, et al. Aberrant expansion of segmented filamentous bacteria in IgA-deficient gut. *Proc Natl Acad Sci U S A* 2004;**101**:1981–6.
87. Umesaki Y. Use of gnotobiotic mice to identify and characterize key microbes responsible for the development of the intestinal immune system. *Proc Jpn Acad Ser B Phys Biol Sci* 2014;**90**:313–32.
88. Atarashi K, Tanoue T, Ando M, Kamada N, Nagano Y, Narushima S, et al. Th17 Cell induction by adhesion of microbes to intestinal epithelial cells. *Cell* 2015;**163**:367–80.
89. Platt AM, Mowat AM. Mucosal macrophages and the regulation of immune responses in the intestine. *Immunol Lett* 2008;**119**:22–31.
90. Romagnani S. Regulation of the T cell response. *Clin Exp Allergy* 2006;**36**:1357–66.
91. Groux H, Garra A, Bigler M, Rouleau M, Antonenko S, Vries JE, et al. A CD4+ T-cell subset inhibits antigen-specific T-cell responses and prevents colitis. *Nature* 1997;**389**:737–42.
92. Akdis M. Healthy immune response to allergens: T regulatory cells and more. *Curr Opin Immunol* 2006;**18**:738–44.
93. Geuking MB, Cahenzli J, Lawson MAE, Ng DCK, Slack E, Hapfelmeier S, et al. Intestinal bacterial colonization induces mutualistic regulatory T cell responses. *Immunity* 2011;**34**:794–806.
94. Furusawa Y, Obata Y, Fukuda S, Endo T a, Nakato G, Takahashi D, et al. Commensal microbe-derived butyrate induces the differentiation of colonic regulatory T cells. *Nature* 2013;**504**:446–50.
95. Josefowicz SZ, Niec RE, Kim HY, Treuting P, Chinen T, Zheng Y, et al. Extrathymically generated regulatory T cells control mucosal TH2 inflammation. *Nature* 2012;**482**:395–9.
96. Atarashi K, Tanoue T, Shima T, Imaoka A, Kuwahara T, Momose Y, et al. Induction of colonic regulatory T cells by indigenous *Clostridium* Species. *Science* 2011;**331**:337–41.
97. Round JL, Mazmanian SK. Inducible Foxp3+ regulatory T-cell development by a commensal bacterium of the intestinal microbiota. *Proc Natl Acad Sci U S A* 2010;**107**:12204–9.
98. Inoue Y, Shimojo N. Microbiome/microbiota and allergies. *Semin Immunopathol* 2015;**37**:57–64.
99. Blazquez AB, Berin MC. Microbiome and food allergy. *Trans Res* 2016; 199–203.
100. Bisgaard H, Li N, Bonnelykke K, Krogsgaard L, Stokholm J, Smith B. Reduced diversity of the intestinal microbiota during infancy is associated with increased risk of allergic disease at school age. *J Allergy Clin Immunol* 2011;**128**:646–52.
101. Kalliomäki M, Kirjavainen P, Eerola E, Kero P, Salminen S, Isolauri E. Distinct patterns of neonatal gut microflora in infants in whom atopy was and was not developing. *J Allergy Clin Immunol* 2001;**107**:129–34.
102. Mah KW, Björkstén B, Lee BW, Van Bever HP, Shek LP, Tan TN, et al. Distinct pattern of commensal gut microbiota in toddlers with eczema. *Int Arch Allergy Immunol* 2006;**140**:157–63.
103. Björkstén B, Sepp E, Julge K, Voor T, Mikelsaar M. Allergy development and the intestinal microflora during the first year of life. *J Allergy Clin Immunol* 2001;**108**:516–20.
104. Abrahamsson TR, Jakobsson HE, Andersson AF, Björkstén B, Engstrand L, Jenmalm MC. Low diversity of the gut microbiota in infants with atopic eczema. *J Allergy Clin Immunol* 2012;**129**:434–40.
105. Fujimura KE, Sitarik AR, Havstad S, Lin DL, Levan S, Fadrosch D, et al. Neonatal gut microbiota associates with childhood multisensitized atopy and T cell differentiation. *Nat Med* 2016;**22**:1187–91.
106. Du Toit G, Foong RXM, Lack G. Prevention of food allergy – early dietary interventions. *Allergol Int* 2016;**65**:370–7.
107. Ling Z, Li Z, Liu X, Cheng Y, Luo Y, Tong X, et al. Altered fecal microbiota composition associated with food allergy in infants. *Appl Environ Microbiol* 2014;**80**:2546–54.
108. Azad MB, Konya T, Guttman DS, Field CJ, Sears MR, HayGlass KT, et al. Infant gut microbiota and food sensitization: associations in the first year of life. *Clin Exp Allergy* 2015;**45**:632–43.
109. Tanaka M, Korenori Y, Washio M, Kobayashi T, Momoda R, Kiyohara C, et al. Signatures in the gut microbiota of Japanese infants who developed food allergies in early childhood. *FEMS Microbiol Ecol* 2017 [in press].
110. Bunyavanich S, Shen N, Grishin A, Wood R, Burks W, Dawson P, et al. Early-life gut microbiome composition and milk allergy resolution. *J Allergy Clin Immunol* 2016;**138**:1122–30.
111. Majkowska-Wojciechowska B, Pelka J, Korzon L, Kozłowska A, Kaczala M, Jarzebska M, et al. Prevalence of allergy, patterns of allergic sensitization and allergy risk factors in rural and urban children. *Allergy* 2007;**62**:1044–50.
112. Von Mutius E, Braun-Fahrlander C, Schierl R, Riedler J, Ehlermann S, Maisch S, et al. Exposure to endotoxin or other bacterial components might protect against the development of atopy. *Clin Exp Allergy* 2010;**30**:1230–4.
113. Debarry J, Hanuszkiewicz A, Stein K, Holst O, Heine H. The allergy-protective properties of *Acinetobacter lwoffii* F78 are imparted by its lipopolysaccharide. *Allergy* 2010;**65**:690–7.
114. Nakayama J, Kobayashi T, Tanaka S, Korenori Y, Tateyama A, Sakamoto N, et al. Aberrant structures of fecal bacterial community in allergic infants profiled by 16S rRNA gene pyrosequencing. *FEMS Immunol Med Microbiol* 2011;**63**:397–406.
115. Vatanen T, Kostic AD, D'Hennezel E, Siljander H, Franzosa EA, Yassour M, et al. Variation in microbiome LPS immunogenicity contributes to autoimmunity in humans. *Cell* 2016;**165**:842–53.
116. Prescott SL, Pawankar R, Allen KJ, Campbell DE, Sinn JK, Fiocchi A, et al. A global survey of changing patterns of food allergy burden in children. *World Allergy Organ J* 2013;**6**:21.
117. Kirjavainen PV, Apostolou E, Arvola T, Salminen SJ, Gibson GR, Isolauri E. Characterizing the composition of intestinal microflora as a prospective treatment target in infant allergic disease. *FEMS Immunol Med Microbiol* 2001;**32**:1–7.
118. He F, Ouwehand AC, Isolauri E, Hashimoto H, Benno Y, Salminen S. Comparison of mucosal adhesion and species identification of bifidobacteria isolated from healthy and allergic infants. *FEMS Immunol Med Microbiol* 2001;**30**:43–7.
119. Shimojo N, Suzuki S, Campos E, Kohno Y. Effects of probiotics on the immune system and allergic diseases. *Allergol Int* 2005;**54**:515–20.
120. Sandin A, Bråbäck L, Norin E, Björkstén B. Faecal short chain fatty acid pattern and allergy in early childhood. *Acta Paediatr* 2009;**98**:823–7.
121. Arrieta M, Stiemsma LT, Dimitriu PA, Thorson L, Russell S, Yurist-Doutsch S, et al. Early infancy microbial and metabolic alterations affect risk of childhood asthma. *Sci Trans Med* 2015;**7**:152.
122. Chen C, Chen K, Kong M, Chang H, Huang J. Alterations in the gut microbiotas of children with food sensitization in early life. *Pediatr Allergy Immunol* 2016;**27**:254–62.
123. Ismail IH, Boyle RJ, Licciardi PV, Oppedisano F, Lahtinen S, Robins-Browne RM, et al. Early gut colonization by *Bifidobacterium breve* and *B. catenulatum* differentially modulates eczema risk in children at high risk of developing allergic disease. *Pediatr Allergy Immunol* 2016;**27**:838–46.