

REVIEW

Microbiota and dietary interactions – an update to the hygiene hypothesis?

R. Frei^{1,2}, R. P. Lauener^{2,3}, R. Cramer¹ & L. O'Mahony¹

¹Swiss Institute of Allergy and Asthma Research, University of Zurich, Davos, Switzerland; ²Christine Kühne-Center for Allergy Research and Education, Zurich, Switzerland; ³Hochgebirgsklinik Davos-Wolfgang, Davos, Switzerland

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Correspondence

Dr. Liam O'Mahony, SIAF, Obere Strasse
22, 7270 Davos Platz, Switzerland.
Tel.: 41 81 4100853
Fax: 41 81 4100840
E-mail: liam.omahony@siaf.uzh.ch

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Abstract

The dramatic increase in the incidence and severity of allergy and asthma has been proposed to be linked with an altered exposure to, and colonization by, micro-organisms, particularly early in life. However, other lifestyle factors such as diet and physical activity are also thought to be important, and it is likely that multiple environmental factors with currently unrecognized interactions contribute to the atopic state. This review will focus on the potential role of microbial metabolites in immunoregulatory functions and highlights the known molecular mechanisms, which may mediate the interactions between diet, microbiota, and protection from allergy and asthma.

Atopic allergic sensitization is characterized by inappropriate immune activation resulting in high levels of specific IgE directed against a range of environmental antigens and animal proteins, leading to allergic disorders including asthma,

rhinitis, and atopic dermatitis (1). These disorders affect 15–25% of Western populations, and their prevalence has increased dramatically over the last few decades. While a number of hypotheses have been proposed to explain this phenomenon, it is clear that there is a strong environmental influence on the incidence and severity of these disorders. Originally, the hygiene hypothesis proposed that allergic diseases were prevented by early infectious exposure introduced by contact with older siblings (2). In support of this hypothesis, a meta-analysis of published articles suggested that early-life *Mycobacterium bovis* Bacillus Calmette–Guerin (BCG) exposure has an inhibitory effect on the development of asthma (3). However, other studies have shown no clear protective effect associated with early-life infections, while additional studies propose that certain infections can actually increase the risk of, and exacerbate, pre-existing asthma (4–10).

In light of these apparently conflicting findings, the hygiene hypothesis has expanded beyond the sole involvement of pathogenic microbes to include interactions of the host with a complex pattern of microbial stimuli. Consistently, a series of studies have shown that children who were in close contact with farm animals, early in life, exhibited significantly lower levels of allergy and asthma in later life (11–16). In addition, the host microbiota (which includes commensal and symbiotic

Abbreviations

APC antigen-presenting cell; BCG Bacillus Calmette–Guerin; CLA conjugated linoleic acid; CLR C-type lectin receptor; DC dendritic cell; DC-SIGN DC-specific intercellular adhesion molecule-grabbing nonintegrin receptor; DHA docosahexaenoic acid; GABA γ -amino butyric acid; Gal Galectin; GPCR G protein-coupled receptor; IDO indoleamine 2,3-dioxygenase; iNOS inducible nitric oxide synthase; ITIM immunoreceptor tyrosine-based inhibitory motif; JNK c-Jun NH(2)-terminal kinase; LCFA long-chain fatty acid; lcfOS long-chain fructo-oligosaccharide; LPS lipopolysaccharide; MCP monocyte chemotactic protein; NGF nerve growth factor; PBMC peripheral blood mononuclear cell; PGE2 prostaglandin E2; PPAR peroxisome proliferator-activated receptor; PRR pattern-recognition receptor; RXR retinoic X receptor; SCFA short-chain fatty acid; scGOS short-chain galacto-oligosaccharide; SHP src homology region 2 domain-containing phosphatase; Siglec Sialic acid-binding immunoglobulin-like lectin; SOCS suppressor of cytokine signaling; T_H T-helper cell; TIM T-cell immunoglobulin mucin; TLR toll-like receptor; T_{reg} T-regulatory cell.

microbes) has been demonstrated to be essential for full immunological development and function in a range of experimental models, in particular germ-free and gnotobiotic animals.

The mechanisms underpinning the hygiene hypothesis were thought to include the polarizing effect of pathogenic microbes on the induction of type 1 T-helper (T_H1) cell responses, thereby inhibiting the type 2 T-helper (T_H2) cell-biased cytokine response associated with the induction of allergy. Understanding of the potential mechanisms has also evolved in the context of altered dendritic cell (DC) and T-regulatory (T_{reg}) cell activity, which is now considered to be a critical feature of the immune dysregulation associated with allergy (17, 18). The list of microbial factors that impact immune cell signaling is growing daily and includes pattern-recognition receptor (PRR) ligands and metabolic products. However, the microbiota is extremely versatile with regard to its ability to utilize a variety of dietary substrates, and the interaction between microbes, diet, and immunoregulation has been poorly described thus far. This review will focus on the influence of the microbiota on immunoregulatory mechanisms and, in particular, the immunoregulatory compounds that are generated by microbial metabolism of dietary factors (Table 1).

Microbial effects on allergy and asthma

It is now widely accepted that the microbiota is important for optimal host development and for ongoing immune homeostasis (19–21). The microbiota aids in the digestion of

foods and nutrient absorption, protects against colonization by pathogens, degrades mucin, and promotes the differentiation of epithelial cells and mucosal-associated lymphoid tissue. In addition, the composition and metabolic activity of the microbiota have profound effects on the induction of immune tolerance. Accumulating evidence suggests that certain bacterial strains may provide protective signals, while other bacterial strains may stimulate aggressive and damaging immune responses. Thus, the activity of the mammalian immune system seems to be governed by the balance between symbiotic and pathogenic factors derived from our microbial inhabitants (22–24). This raises the possibility that dysbiosis can lead to inappropriate inflammatory responses and may contribute to the dramatic increase in several immune-mediated disorders observed in recent decades, including allergy and asthma. However, it remains unclear whether dysbiosis is the cause or an epiphenomenon of the disease.

There is significant interest in the deliberate administration of microbes or microbial metabolites for the treatment of aberrant inflammatory activity (25–30). The protective effects associated with these microbes are probably mediated by multiple mechanisms involving epithelial cells, DCs, and T cells. Despite encouraging animal model data, human clinical studies examining the effectiveness of therapeutic microbes for the prevention or treatment of allergic disease have not been conclusive thus far. A review of 13 randomized placebo-controlled trials concluded that certain, but not all, microbes could have utility particularly when administered early in life (31). In particular, infants at high risk of atopy

Table 1 Evidence for the interaction between microbial metabolites and the immune system

Dietary component	Microbial metabolite	Host effect	Assessed	References
Carbohydrates	Polysaccharides	IL-10	<i>In vitro</i> /Animal studies	(87, 88, 90, 95, 98)
		IL-12		(90)
		Superoxide		(90)
		Migration		(90, 91)
		Adhesion		(90, 91)
		Mast cell degranulation		(92, 101)
		Apoptosis		(91, 92)
Nondigestible carbohydrates	Short-chain fatty acid (SCFA)	Prostaglandin E2 (PGE2)	<i>In vitro</i> /Animal studies	(103, 110)
		Inflammation		(103, 104)
		dendritic cell differentiation		(111, 112)
		IL-10		(103, 110)
		iNOS		(103, 110)
		Mast cell degranulation		(113, 114)
		Inflammation		(115–118)
Prebiotics, Sugar, Antioxidants, SCFA	Long-chain fatty acid	NO	<i>In vitro</i> /Animal and human clinical studies	(75, 121)
		PGE2, Leukotriene B4		(75, 121)
		IL-10		(75, 121)
		IgE		(121)
		IgA		(121)
		Maturation		(122, 123)
		Activation		(122, 123)
Aldehyde, Ketone, Fermentable carbohydrates, Oxygen	Biogenic amines	Polarization	<i>In vitro</i> /Animal and human clinical studies	(122, 123)
		Effector function		(122, 123)
		Inflammation		(122, 123)
		Mucus production		(124, 125)

who received *Lactobacillus rhamnosus* GG developed atopic dermatitis significantly less frequently during the first 2 years of life. However, prenatal treatment alone failed to prevent atopic eczema during the first year of life (32). A more recent study, which used a combination of three bacterial strains, demonstrated that prenatal and postnatal supplementation was effective in preventing the development of atopic eczema in infants at high risk of allergy during the first year of life (33). The use of microbes for the treatment of allergic disease is less promising. A Cochrane meta-analysis concluded that the microbes tested to date are not effective for the treatment of eczema (34). In addition, clinical data on the use of microbes in the treatment of asthma and allergic rhinitis are conflicting. One issue with the selection of immunoregulatory microbes is that the most suitable bacterial strain may not have been chosen, as not all probiotic strains exert the same effects *in vivo* compared with the results obtained *in vitro*. In addition, the metabolic activity of a bacterial strain is significantly influenced by the availability of suitable substrates, thereby leading to the secretion of varying quantities of different metabolites. Dietary factors are not usually considered in probiotic clinical studies, which may contribute to the inconsistent effects observed thus far.

Microbe–host immunoregulatory cellular mechanisms

In order to ensure the induction of tolerance and protective immunity, discriminative responses are required to commensals in comparison with pathogens, respectively. For example, segmented filamentous bacteria can promote the differentiation of T-helper 17 (T_H17) cells, while certain *Bifidobacterium*, *Lactobacillus*, and *Clostridium* species can promote T_{reg} development (24, 35–37). At body surfaces that come into contact with bacteria, a number of cell types including DCs, lymphocytes, and epithelial cells are required to maintain immunological homeostasis and tolerance (38).

Dendritic cells are professional antigen-presenting cells (APCs), which are in close contact with microbes and are responsible for presenting microbial, environmental, and dietary antigens to the adaptive immune system, thereby influencing the polarization of the adaptive response via cytokine and metabolite production. Dendritic cells are proposed to be an important cellular component of the hygiene hypothesis, as suggested by murine studies. Dendritic cells isolated from mice, which were infected with *Schistosoma japonicum*, *Chlamydia muridarum*, or BCG, were able to inhibit allergic airway inflammation following adoptive transfer (39–41). Depending on the specific infection, DCs secreted IL-10, IL-12, or both, suggesting that tolerogenic and T_H1-polarizing DCs, induced by an infection, efficiently inhibit the induction of T_H2 allergic polarization. Dendritic cells recognize different microbial structures and components via their expression of PRRs, which program DC gene expression and subsequent T-cell polarization (42). For example, Toll-like receptor (TLR)-2 recognition of zymosan results in the secretion of retinoic acid and IL-10 leading to Foxp3 induction, while dectin-1 activation by zymosan leads to IL-23 secretion and T_H17 induction (43). Recent findings on the role of PRR

signaling in mucosal homeostasis have emphasized the delicate balance between different PRR functions and revealed that defective PRR signaling can result in inflammation and atopic sensitization (44, 45). Toll-like receptor-2 knock-out animals are more sensitive to dextran sodium sulfate-induced colitis, while TLR-2 gene variants are associated with disease phenotype in patients with inflammatory bowel disease. Indeed, TLR-2 has been demonstrated to promote Foxp3 expression in response to intestinal microbes in murine models (46, 47). In addition, cross-talk between TLR-2/6, DC-specific intercellular adhesion molecule-grabbing nonintegrin receptor (DC-SIGN), and TLR-9 activation on human DCs, in response to a commensal microbe, is responsible for inducing high levels of IL-10 secretion and lymphocyte Foxp3 expression (48). Toll-like receptor 3, but not TLR4, is responsible for IFN- λ secretion by intestinal DCs, which has been shown to play an important role in the maintenance of oral tolerance (49).

The primary function of epithelial cells is to provide a physical barrier between the external environment and the underlying body organs. In addition, epithelial cells provide immunoregulatory signals that can influence DC and lymphocyte activation. Epithelial cells express PRRs and therefore can respond to the presence of different microbial species. For example, lipoteichoic acid released by *Staphylococcus epidermidis* (a cutaneous commensal microbe) inhibits both inflammatory cytokine release from keratinocytes and inflammation triggered by injury through a TLR-2-dependent process (50). Intestinal epithelial cells regulate DC, T- and B-cell functions through the secretion of immunomodulatory molecules, such as chemokines, retinoic acid, thymic stromal lymphopoietin, and TGF- β (51–53). The presence of certain commensal microbes can influence the production of chemokines and regulatory factors by epithelial cells. *Lactobacillus* and *Bifidobacterium* strains have been shown to inhibit intestinal epithelial cell NF- κ B activation and chemokine secretion to a range of proinflammatory and pathogenic stimuli, including flagellin, TNF- α , *Salmonella enterica* serovar Typhimurium, and *Clostridium difficile* (54–56). Multiple mechanisms are thought to be involved in the direct inhibition of epithelial proinflammatory responses, which include the secretion of nerve growth factor (NGF), the prevention of the degradation of the NF- κ B inhibitor I κ B- α , or the nuclear export of the p65 subunit of NF- κ B in a peroxisome proliferator-activated receptor (PPAR) γ -dependent manner (54, 57, 58).

A common feature of immunoregulatory microbes, which is being increasingly reported, is their ability to induce T_{reg} cells. For example, encounters with a mixture of commensal microbes (VSL#3 probiotic cocktail) within the murine gut have been shown to drive the development of mucosal T_{reg} cells, which is associated with the attenuation of inflammation in a murine model of colitis (59). In addition, the consumption of a *Bifidobacterium infantis* strain promotes T_{reg}-cell conversion and protects against lipopolysaccharide (LPS)-induced NF- κ B activation *in vivo*, while *Lactobacillus reuteri* induces T_{reg} cells that protect against an allergic airway response in mice (37, 60). T_{reg} cells are derived from

the thymus but may also be induced in peripheral organs, including the gut mucosa (61, 62). CD103⁺ DCs within the mucosa are largely responsible for the conversion of T_{reg} cells via TGF- β and retinoic acid-dependent processes (63, 64). The conversion is likely driven by gastrointestinal-specific environmental factors associated with the presence of large numbers of commensal organisms. Indeed, we have recently reported that retinoic acid-metabolizing enzymes can be up-regulated in myeloid, but not in plasmacytoid, DCs following *in vitro* incubation with a *Bifidobacterium infantis* bacterial strain (48). However, it is unlikely that all commensal microbes are equally effective at inducing T_{reg} cells *in vivo*. A recent study comparing multiple commensal organisms (*Bifidobacterium longum* AH1206, *Bifidobacterium breve* AH1205, and *Lactobacillus salivarius* AH102) has shown that *Bifidobacterium longum* AH1206 induced T_{reg} cells and was also able to protect against respiratory allergic inflammation (36). The other bacterial strains did not effectively induce T_{reg} cells and were not able to protect against allergic inflammation. Therefore, the induction of T_{reg} cells may be a critical characteristic of a healthy microbiota, which is protective against the development of aberrant immunological reactivity to potential allergens. In addition to using live microbes for the treatment of allergy, another exciting approach is to identify the microbial factor(s) responsible for the beneficial effect and to use these isolated factor(s) alone. For example, polysaccharide A derived from *Bacteroides fragilis* promotes an appropriate T_H1/T_H2 balance in germ-free mice following presentation by mucosal DCs and protects against colitis in an animal model via IL-10-secreting CD4⁺ T cells (29, 65). The continued identification of new microbial compounds, which induce tolerogenic DCs and T_{reg} activity, will undoubtedly lead to novel therapeutic candidate molecules for assessment in clinical studies.

Dietary factors that influence microbial metabolism

A wide range of complex oligosaccharides, also called fiber, are not digested by the host, but enter the colon and provide fermentable substrates for the colonic microbiota (66). Historically, diets high in fiber have been shown to diminish the severity of arthritis and inflammatory bowel diseases (67). Certain dietary fibers that promote the growth of *Bifidobacteria* and *Lactobacilli* strains have been identified and these are called prebiotics. Typically, prebiotics are short-chain carbohydrates with a degree of polymerization between two and sixty, such as polydextrose, inulin, fructo-oligosaccharides, galacto-oligosaccharides, and soybean oligosaccharides (68–71). Bacterial metabolic processing of these oligosaccharides leads to the generation of novel polysaccharide structures, in addition to the secretion of short-chain fatty acids (SCFAs). *Bifidobacteria* utilize the 'bifid shunt' catabolic pathway to produce SCFAs and ATP. Before entering the 'bifid shunt' pathway, oligosaccharides are processed into constituent carbohydrates by glycosyl hydrolases (72). The catabolic pathway uses fructose-6-phosphate phosphoketolase to produce phosphoenolpyruvate that is degraded to pyruvate, the primary metabolite for SCFAs production. Pyruvate is fur-

ther metabolized to acetyl-CoA leading to the formation of acetate and butyrate (73). Depending on the specific polysaccharides, different SCFAs can be produced. Pectin and xylan fermentation primarily leads to acetate generation. Arabino-galactan breakdown results in acetate and propionate synthesis, while starch was shown to be metabolized into butyrate (66). SCFAs are either secreted or incorporated in newly synthesized fatty acids within cellular structures.

In addition to SCFAs, long-chain fatty acids (LCFAs) are produced by bacterial metabolism within the gut. Long-chain fatty acids such as omega-3 fatty acids or conjugated linoleic acids (CLAs) have been reported to have beneficial effects on cancer, cardiovascular disease, diabetes, obesity, bone disorders, and inflammatory diseases as well as to have anti-oxidative and growth-promoting properties (67, 74). Meat and milk products derived from ruminants are a traditional source of CLAs. In the rumen, CLAs are produced from free linoleic acid by bacterial isomerase activity (75). Antioxidants, sugar, and certain prebiotic compounds as well as the sodium salt of SCFAs had a positive impact on the CLAs produced by *Bifidobacteria* in the intestine of mammals (66, 74). Certain probiotic bacteria are able to produce CLAs from linoleic acid, vaccenic acid, or hydroxyl fatty acids (75).

Biogenic amines are synthesized by decarboxylation of amino acids or by amination and transamination of aldehydes and ketones. Biogenic amines such as γ -amino butyric acid (GABA) or histamine can be synthesized by bacteria (76). Enzymes that are able to generate biogenic amines are found in microbes, plants, and animal metabolic processes. In bacteria, the expression and activity of amino acid decarboxylases is enhanced in acidic environments, such as in the stomach. This leads to a local increase in pH around the bacteria and protects it from the acidic, chloride-rich environment (76, 77). Furthermore, expression and activity of decarboxylases in bacteria is regulated by the presence of fermentable carbohydrates and oxygen, by the redox potential of the medium, by the temperature, and by the sodium chloride concentration (77).

Immunoregulatory mechanisms associated with bacterial metabolites

The dietary factors and their known effects on host immunoregulation are summarized in Fig. 1. Nondigestible carbohydrates may influence the immune response, independently of stimulating the growth of the microbial flora in the gut, potentially through two immunoregulatory mechanisms. The generation of carbohydrate structures, which bind PRRs, may influence immune regulatory activity, while SCFAs may also bind receptors on gut-associated cells.

Polysaccharides

Carbohydrate binding proteins are increasingly being shown to regulate immune reactivity and immune tolerance mechanisms. Lectins are carbohydrate binding proteins and are expressed in virtually all animals and plants. The lectin family is categorized based on the structure and

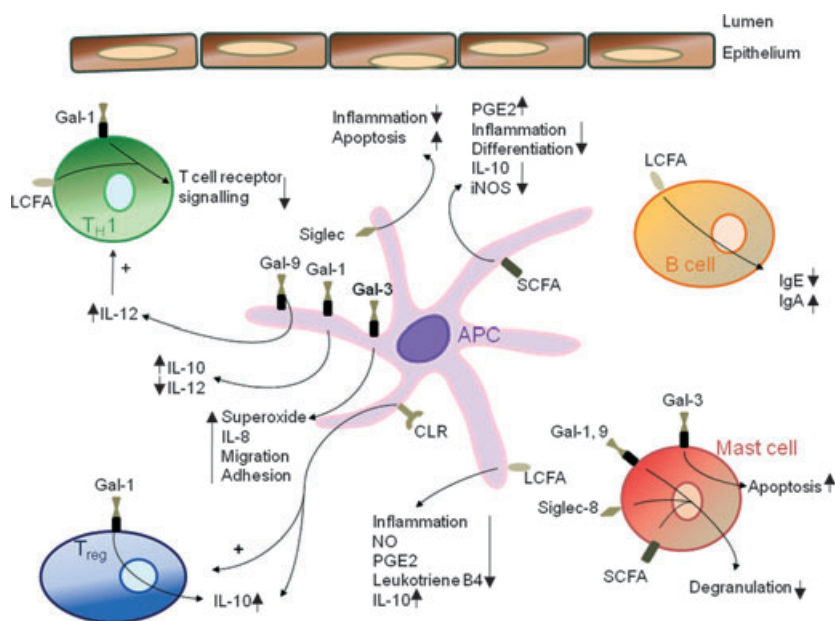


Figure 1 Microbial metabolites such as lectin-binding carbohydrates, Short-chain fatty acids, and long-chain fatty acids influence immune responses such as inflammatory responses, immune

regulation, cell migration and adhesion, and eicosanoid production in antigen-presenting cells, mast cells, T-helper cells, and B cells.

function (78, 79). Many of the C-type lectins (CLRs) and sialic acid-binding immunoglobulin-like lectins (Siglec) are membrane bound, while the galectins are secreted. All lectins contain one or more carbohydrate-recognition domains (80). They are involved in the activation or regulation of TLR-dependent or independent innate gene expression and antigen presentation. Lectins recognize various carbohydrate structures of pathogens, such as parasites, viruses, bacteria, and fungi. Additionally, lectins are further involved in translating glycan-encoded information of host cells into an effector immune function (81–84). Binding of lectins to their ligands can positively or negatively influence the priming of the immune system and induction of tolerance (81).

C-type lectin receptors recognize monosaccharides, such as mannose, fucose, galactose, or *N*-acetylgalactosamine and more complex carbohydrate profiles of various pathogens or of host cells by their C-type carbohydrate-recognition domains leading to internalization into intracellular compartments of DCs resulting in the processing and presentation of the antigenic peptides on MHC class I and II molecules (82–84). In general, targeting of CLRs on immature DCs in the absence of TLR stimulation does not lead to DC maturation but to peripheral tolerance and to induction of T_{reg} cells, which is one mechanism exploited by tumor cells and some viruses to dampen an immune response (84–86). So far, 15 CLRs have been identified in DCs and macrophages. One well-described CLR is DC-SIGN, a receptor for mannose-containing particles that binds HIV, Ebola, or mycobacteria (82, 83). Targeting DC-SIGN on human DCs activates the serine and threonine kinase Raf-1 leading to the acetylation of the NF- κ B subunit p65 and increased IL-10 transcription (87). Interestingly, it has been shown that a sugar-modified

antigen can induce oral tolerance in a mouse model of food allergy. This process seems to be dependent on IL-10, induced via the CLR SIGNR1 on DCs in the gastrointestinal lamina propria (88). Another mannose receptor is langerin (CD207), selectively expressed on Langerhans cells (82). Dectin-1 plays a crucial role in regulating immune responses against fungi via recognition of β -glucans (82). The mannose receptor (CD206) has been described to recognize various airborne allergens leading to induction of a T_H2 response by the inhibition of indoleamine 2,3-dioxygenase (IDO) activity (89).

S-type lectins comprise a group of 15 different galectins (82), which require a reduced thiol group for full function (78, 79). Galectins are soluble lectins that bind to multiple glycosylated structures on host cells (90). Galectins are involved in cell signaling, cell adhesion, chemotaxis, lipid raft stabilization, and cell apoptosis (91). They act as receptors for *N*-acetyllactosamine and are expressed by activated T cells and B cells, eosinophils, basophils, thymic and intestinal epithelia cells and are upregulated in activated macrophages and T_{reg} cells (80). Variations in the activity of glycosyltransferases and glycosidases modulate the ligand density for galectins and thereby the activation of T cells, cytokine secretion, or apoptosis (82). Interestingly, galectin-9 and TLR-9 have been shown to mediate the protective effect of a diet containing a probiotic bacterial strain combined with a mixture of short-chain galacto-oligosaccharides (scGOS) and long-chain fructo-oligosaccharides (lcFOS) in a food allergy model (91). Galectin-3 induces neutrophil and macrophage migration and adhesion to endothelia and IL-8 secretion and activates NADPH oxidase leading to superoxide production (90). Galectin-1 modulates phagocytosis and antigen

presentation of macrophages by regulating Fc γ receptor 1 and MHC class II. Furthermore, IL-12 production is inhibited and IL-10 production increased in galectin-1-stimulated macrophages and DCs (90). Mast cells from galectin-3-deficient mice secreted less histamine and IL-4 upon Fc ϵ RI cross-linking (92). Extracellular galectin-3 induces apoptosis in mast cells, while galectin-1 and -9 reduce their ability to degranulate, possibly due to the inhibition of IgE-antigen complex formation (90). Intestinal epithelial cells express galectin-2, -3, -4, -5, -6, -7, and -9 (91). Galectin-1 has been shown to antagonize T-cell receptor signaling by inducing partial phosphorylation of the T-cell receptor ξ -chain and suppression of IL-2 production. Galectin-1 also blocks CD2 binding of T cells to APCs in the immunological synapse. Furthermore, galectin-1 enhances the secretion of IL-10 by T_{reg} cells. Galectin-3 regulates T-cell receptor activation by binding the T-cell receptor. In addition, binding of galectin-9 to T-cell immunoglobulin mucin (TIM)-3 receptor on DCs induces IL-12 production in a p38-dependent manner and enhances T_H1 response. Depending on the relative concentration of galectin-9, IFN- γ can be induced by T_H1 cells, or galectin-9 can induce the development of T_{reg} cells and suppress T_H17 differentiation in mice (91).

Siglecs are I-type lectins, belong to the immunoglobulin superfamily, and recognize sialic acid attached to the terminal regions of cell-surface glycoconjugates (82, 93–96). They are categorized into two groups (94). Members of the more conserved group are sialoadhesin (Siglec-1/CD169), CD22 (Siglec-2), myelin-associated glycoprotein (Siglec-4), and Siglec-15. They have 25–30% sequence identity and have orthologues in all mammalian species. The group of CD33-related Siglecs share 50–99% sequence identity but their rapid evolution has resulted in differences between mammalian species. Nine human (CD33, Siglec-5 to 11, Siglec-14, and -16) and five murine CD33-related Siglecs (CD33, Siglec-E, -F, -G, and -H) have been identified. (93–96). Many immune cell subsets, except T cells, express at least one Siglec (80, 96, 97). Many Siglecs have intracellular immunoreceptor tyrosine-based inhibitory motifs (ITIM) indicating an inhibitory function that could suppress activation signals. After ligation, the ITIM is phosphorylated enabling the recruitment of the signaling molecules src homology region 2 domain-containing phosphatase (SHP)-1 and -2 tyrosine phosphatase and the suppressor of cytokine signaling (SOCS)-3 (97). In fact, Siglecs dampen immune response activation or induce apoptosis also by enhancing the production of anti-inflammatory cytokines (95, 98, 99). Each Siglec has a unique ligand-specificity profile. Targeting human Siglec-8 or Siglec-F, the mouse homolog, has been shown to regulate eosinophil viability by the induction of apoptosis (100, 101). Furthermore, Fc ϵ RI-dependent histamine and prostaglandin D₂ release was blocked by the engagement of Siglec-8 on mast cells (102).

SCFA

Short-chain fatty acids are ligands for G protein-coupled receptors (GPCRs), such as GPR43 and GPR41. Both fatty acid receptors are involved in metabolism and immunity.

SCFAs, particularly butyrate, seem to exert anti-inflammatory activities by affecting immune cell migration, adhesion, cytokine expression as well as by affecting cellular processes such as proliferation, activation, and apoptosis (103). GPR43 activation has been shown to be necessary for the optimal resolution of certain inflammatory responses, because GPR43-deficient mice showed exacerbated or un-resolving inflammation in the models of colitis, arthritis, and asthma. GPR43 deficiency is related to increased production of inflammatory mediators by immune cells and to increased immune cell recruitment (104). GPR43 is expressed not only on immune cells such as monocytes and neutrophils but also on colonic epithelium, while GPR41 is more widely distributed over tissues but is particularly expressed on adipose tissues, immune and endothelial cells (105). Both receptors couple through G_q and G_{i/o} families. The resulting intracellular pathways activated by these receptors include inositol 1,4,5-trisphosphate generation, intracellular Ca²⁺ release, ERK1/2 and p38 activation, and inhibition of cAMP accumulation (106, 107). Depending on the concentration and type of SCFAs, GPR43 ligand binding can induce or reduce neutrophil migration and cell adhesion to endothelial cells (103, 108, 109). Short-chain fatty acids induce the release of prostaglandin E₂ (PGE₂) in human monocytes and inhibit constitutive monocyte chemotactic protein-1 (MCP-1) production and LPS-induced IL-10 production in human monocytes. Similar activities were observed in human peripheral blood mononuclear cells (PBMCs) with regard to PGE₂, MCP-1, and IL-10 secretion after SCFAs treatment. In addition, SCFAs inhibit LPS-induced I κ B degradation and therefore the production of TNF- α , IFN- γ , IL-6, IL-1 β , and the inducible nitric oxide synthase (iNOS) in human PBMCs (103, 110). These effects seem to be strongest for butyrate, while propionate and acetate increased IFN- γ and IL-10 release. SCFAs inhibit DC differentiation from bone marrow stem cells by the regulation of gene transcription via inhibition of histone deacetylases (111, 112). Butyrate has been shown to inhibit mast cell degranulation and TNF- α production by inhibiting c-Jun NH(2)-terminal kinase (JNK) and phospholipase D (113). Furthermore, SCFAs inhibited antigen-induced contraction of guinea pig tracheal rings and colon and histamine release by rat peritoneal mast cells (114).

LCFA

Long-chain fatty acids such as omega-3 fatty acids or CLA possess immunoregulatory activity. The omega-3 fatty acid docosahexaenoic acid (DHA) inhibits p38 MAPK phosphorylation and the degradation of I κ B kinase, following binding to GPR120 and PPARs. GPR120 is a physiological receptor for omega-3 fatty acids in macrophages and adipocytes. Ligand binding mediates anti-inflammatory and insulin-sensitizing effects (115). Binding of GPR120 by omega-3 fatty acids results in the internalization of GPR120, which binds β -arrestin 2 and sequesters TAB 1, a molecule crucial for TLR- or TNF- α -mediated induction of inflammation (116). PPARs form a heterodimer with retinoic X receptors

(RXRs), subsequently regulating many target genes upon activation via transrepression (117). PPAR ligands suppress the expression of various inflammatory components such as cytokines, chemokines, and costimulatory molecules in macrophages and DCs (118). Omega-3 fatty acids diminished DC-induced T-helper cell activation by influencing antigen presentation and costimulatory molecule signaling (119, 120). Ligand binding to PPARs within T cells reduces the secretion of IFN- γ and IL-17 (118).

There are two mechanisms described by which CLAs influence the immune response. CLAs diminish eicosanoid production leading to the downregulation of PGE2 and leukotriene B4. In addition, CLAs have been shown to bind PPARs. Humans, or guinea pigs, with diets supplemented with CLAs have higher IL-10, lower TNF- α , and lower IFN- γ serum levels. Furthermore, CLAs inhibit LPS-induced TNF- α and NO production (75, 121). Dietary supplementation with CLAs in humans results in higher IgA and decreased IgE serum levels. Furthermore, CLAs increase the immune response toward hepatitis B vaccination and mice display enhanced CD8+ T-cell numbers after feeding with CLAs (121).

Biogenic amines

The biological consequences of biogenic amine secretion *in vivo* by the resident microbiota are largely unknown. Histamine production by bacteria present within decaying fish can result in scombroid food poisoning, which resembles symptoms associated with an allergic response. Histamine has been shown to exert potent immunoregulatory effects and may negatively or positively influence parasitic or bacterial infections (122). A wide range of host immune cells can express histamine receptors, which can selectively recruit the major effector cells into tissue sites and affect their maturation, activation, polarization, and effector functions leading to tolerogenic or pro-inflammatory responses (123). However, the influence of histidine-rich or depleted diets on histamine secretion by the enteric microbiota is currently unknown. Another biogenic amine, which has been shown to be produced by microbes present within certain foods, is GABA (124). Two types of GABA receptors are known. The GABA_A receptor subtype forms a part of a ligand-gated ion channel complex, and the GABA_B receptor subtype is a GPCR. Interestingly, the GABA signaling pathway has been shown to be important for the stimulation of mucus

production from airway epithelial cells, in both mouse models of respiratory allergy and allergen-challenged asthma patients (125). The contribution of enteric microbes to the systemic GABA pool, via the metabolism of glutamate *in vivo*, is unknown and requires further investigation.

Concluding remarks

While it is clear that the microbiota significantly influences host immune maturation and immune activity, the molecular basis for these immunomodulatory mechanisms is only beginning to be elucidated. The presence of certain bacterial species or strains seems to be important, potentially because of direct interactions with the host (e.g., via PRR activation) or via their metabolic activity *in vivo* (e.g., SCFAs generation). Thus, care should be exercised in the selection of immunoregulatory microbes for administration in human studies as it is likely that not all microbes are equally effective and dietary factors may significantly influence the production of immunoregulatory metabolites. In addition, significant effort needs to be focused on the elucidation of the microbiota-associated molecular pathways that are impacted by dietary factors so that rationale prevention and treatment strategies can be formulated, which will include matching essential microbes and dietary components. Specific projects mining the microbiota for metabolites and ligands that modulate host immune function will likely lead to a new class of immunotherapeutic agents with relevance to a variety of inflammatory states, including allergy and asthma. Lastly, the hygiene hypothesis should now be updated to include dietary factors and the interaction of dietary factors with the microbiota.

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Conflicts of interest

Liam O'Mahony is a consultant to Alimentary Health Ltd. R.F. and R.C. have no conflicts of interests to declare.

References

1. Kay AB. Allergy and allergic diseases. First of two parts. *N Engl J Med* 2001;**344**:30–37.
2. Strachan DP. Hay fever, hygiene, and household size. *BMJ* 1989;**299**:1259–1260.
3. El-Zein M, Parent ME, Benedetti A, Rousseau MC. Does BCG vaccination protect against the development of childhood asthma? A systematic review and meta-analysis of epidemiological studies. *Int J Epidemiol* 2010;**39**:469–486.
4. Ramsey CD, Celedon JC. The hygiene hypothesis and asthma. *Curr Opin Pulm Med* 2005;**11**:14–20.
5. Linneberg A, Ostergaard C, Tvede M, Andersen LP, Nielsen NH, Madsen F et al. IgG antibodies against microorganisms and atopic disease in Danish adults: the Copenhagen Allergy Study. *J Allergy Clin Immunol* 2003;**111**:847–853.
6. Rennie DC, Lawson JA, Kirychuk SP, Paterson C, Willson PJ, Senthilselvan A et al. Assessment of endotoxin levels in the home and current asthma and wheeze in school-age children. *Indoor Air* 2008;**18**:447–453.
7. Dahl ME, Dabbagh K, Liggitt D, Kim S, Lewis DB. Viral-induced T helper type 1 responses enhance allergic disease by effects on lung dendritic cells. *Nat Immunol* 2004;**5**:337–343.
8. Park JH, Gold DR, Spiegelman DL, Burge HA, Milton DK. House dust endotoxin

- and wheeze in the first year of life. *Am J Respir Crit Care Med* 2001;**163**:322–328.
9. Hogg JC. Childhood viral infection and the pathogenesis of asthma and chronic obstructive lung disease. *Am J Respir Crit Care Med* 1999;**160**(5 Pt 2):S26–S28.
 10. Cramer C, Link E, Horster M, Koletzko S, Bauer CP, Berdel D et al. Elder siblings enhance the effect of filaggrin mutations on childhood eczema: results from the 2 birth cohort studies LISApus and GINIplus. *J Allergy Clin Immunol* 2010;**125**:1254–1260.
 11. Araujo MI, Hoppe B, Medeiros M Jr, Alcantara L, Almeida MC, Schriefer A et al. Impaired T helper 2 response to aeroallergen in helminth-infected patients with asthma. *J Infect Dis* 2004;**190**:1797–1803.
 12. 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* 2000;**30**:1230–1234.
 13. Debarry J, Garn H, Hanuszkiewicz A, Dickgreber N, Blumer N, von Mutius E et al. Acinetobacter Iwoffii and Lactococcus lactis strains isolated from farm cowsheds possess strong allergy-protective properties. *J Allergy Clin Immunol* 2007;**119**:1514–1521.
 14. Ege MJ, Bieli C, Frei R, van Strien RT, Riedler J, Ublagger E et al. Prenatal farm exposure is related to the expression of receptors of the innate immunity and to atopic sensitization in school-age children. *J Allergy Clin Immunol* 2006;**117**:817–823.
 15. Roduit C, Wohlgensinger J, Frei R, Bitter S, Bieli C, Loeliger S et al. Prenatal animal contact and gene expression of innate immunity receptors at birth are associated with atopic dermatitis. *J Allergy Clin Immunol* 2011;**127**:179–185.
 16. Ege MJ, Mayer M, Normand AC, Genuneit J, Cookson WO, Braun-Fahrlander C et al. Exposure to environmental microorganisms and childhood asthma. *N Engl J Med* 2011;**364**:701–709.
 17. Romagnani S. The increased prevalence of allergy and the hygiene hypothesis: missing immune deviation, reduced immune suppression, or both? *Immunology* 2004;**112**:352–363.
 18. Akdis M, Akdis CA. Therapeutic manipulation of immune tolerance in allergic disease. *Nat Rev Drug Discov* 2009;**8**:645–660.
 19. O'Hara AM, Shanahan F. The gut flora as a forgotten organ. *EMBO Rep* 2006;**7**:688–693.
 20. Shanahan F. Gut microbes: from bugs to drugs. *Am J Gastroenterol* 2010;**105**:275–279.
 21. Lee YK, Mazmanian SK. Has the microbiota played a critical role in the evolution of the adaptive immune system? *Science* 2010;**330**:1768–1773.
 22. Grainger JR, Hall JA, Bouladoux N, Oldenhove G, Belkaid Y. Microbe-dendritic cell dialog controls regulatory T-cell fate. *Immunol Rev* 2010;**234**:305–316.
 23. Littman DR, Rudensky AY. Th17 and regulatory T cells in mediating and restraining inflammation. *Cell* 2010;**140**:845–858.
 24. Feng T, Elson CO. Adaptive immunity in the host-microbiota dialog. *Mucosal Immunol* 2011;**4**:15–21.
 25. van der Kleij H, O'Mahony C, Shanahan F, O'Mahony L, Bienenstock J. Protective effects of Lactobacillus rhamnosus [corrected] and Bifidobacterium infantis in murine models for colitis do not involve the vagus nerve. *Am J Physiol Regul Integr Comp Physiol* 2008;**295**:R1131–R1137.
 26. Sheil B, McCarthy J, O'Mahony L, Bennett MW, Ryan P, Fitzgibbon JJ et al. Is the mucosal route of administration essential for probiotic function? Subcutaneous administration is associated with attenuation of murine colitis and arthritis. *Gut* 2004;**53**:694–700.
 27. McCarthy J, O'Mahony L, O'Callaghan L, Sheil B, Vaughan EE, Fitzsimons N et al. Double blind, placebo controlled trial of two probiotic strains in interleukin 10 knockout mice and mechanistic link with cytokine balance. *Gut* 2003;**52**:975–980.
 28. O'Mahony L, Feeney M, O'Halloran S, Murphy L, Kiely B, Fitzgibbon J et al. Probiotic impact on microbial flora, inflammation and tumour development in IL-10 knockout mice. *Aliment Pharmacol Ther* 2001;**15**:1219–1225.
 29. Mazmanian SK, Round JL, Kasper DL. A microbial symbiosis factor prevents intestinal inflammatory disease. *Nature* 2008;**453**:620–625.
 30. Grabig A, Paclik D, Guzy C, Dankof A, Baumgart DC, Erckenbrecht J et al. Escherichia coli strain Nissle 1917 ameliorates experimental colitis via toll-like receptor 2- and toll-like receptor 4-dependent pathways. *Infect Immun* 2006;**74**:4075–4082.
 31. Betsi GI, Papadavid E, Falagas ME. Probiotics for the treatment or prevention of atopic dermatitis: a review of the evidence from randomized controlled trials. *Am J Clin Dermatol* 2008;**9**:93–103.
 32. Boyle RJ, Ismail IH, Kivivuori S, Licciardi PV, Robins-Browne RM, Mah LJ et al. Lactobacillus GG treatment during pregnancy for the prevention of eczema: a randomized controlled trial. *Allergy* 2011;**66**:509–516.
 33. Kim JY, Kwon JH, Ahn SH, Lee SI, Han YS, Choi YO et al. Effect of probiotic mix (Bifidobacterium bifidum, Bifidobacterium lactis, Lactobacillus acidophilus) in the primary prevention of eczema: a double-blind, randomized, placebo-controlled trial. *Pediatr Allergy Immunol* 2010;**21**(2 Pt 2):e386–e393.
 34. Boyle RJ, Bath-Hextall FJ, Leonardi-Bee J, Murrell DF, Tang ML. Probiotics for treating eczema. *Cochrane Database Syst Rev* 2008;**4**:CD006135.
 35. 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**:337–341.
 36. Lyons A, O'Mahony D, O'Brien F, MacSharry J, Sheil B, Ceddia M et al. Bacterial strain-specific induction of Foxp3+ T regulatory cells is protective in murine allergy models. *Clin Exp Allergy* 2010;**40**:811–819.
 37. Karimi K, Inman MD, Bienenstock J, Forsythe P. Lactobacillus reuteri-induced regulatory T cells protect against an allergic airway response in mice. *Am J Respir Crit Care Med* 2009;**179**:186–193.
 38. Atarashi K, Umesaki Y, Honda K. Microbial influence on T cell subset development. *Semin Immunol* 2011;**23**:146–153.
 39. Han X, Wang S, Fan Y, Yang J, Jiao L, Qiu H et al. Chlamydia infection induces ICOS ligand-expressing and IL-10-producing dendritic cells that can inhibit airway inflammation and mucus overproduction elicited by allergen challenge in BALB/c mice. *J Immunol* 2006;**176**:5232–5239.
 40. Bilenki L, Gao X, Wang S, Yang J, Fan Y, Han X et al. Dendritic cells from mycobacteria-infected mice inhibits established allergic airway inflammatory responses to ragweed via IL-10- and IL-12-secreting mechanisms. *J Immunol* 2010;**184**:7288–7296.
 41. Liu P, Li J, Yang X, Shen Y, Zhu Y, Wang S et al. Helminth infection inhibits airway allergic reaction and dendritic cells are involved in the modulation process. *Parasite Immunol* 2010;**32**:57–66.
 42. Pulendran B, Tang H, Manicassamy S. Programming dendritic cells to induce T(H)2 and tolerogenic responses. *Nat Immunol* 2010;**11**:647–655.
 43. Manicassamy S, Ravindran R, Deng J, Oluoch H, Denning TL, Kasturi SP et al. Toll-like receptor 2-dependent induction of vitamin A-metabolizing enzymes in dendritic cells promotes T regulatory responses and inhibits autoimmunity. *Nat Med* 2009;**15**:401–409.
 44. Fukata M, Abreu MT. Pathogen recognition receptors, cancer and inflammation in the gut. *Curr Opin Pharmacol* 2009;**9**:680–687.
 45. Tesse R, Pandey RC, Kabesch M. Genetic variations in toll-like receptor pathway

- genes influence asthma and atopy. *Allergy* 2011;**66**:307–316.
46. Round JL, Lee SM, Li J, Tran G, Jabri B, Chatila TA et al. The Toll-like receptor 2 pathway establishes colonization by a commensal of the human microbiota. *Science* 2011;**332**:974–977.
 47. 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**:12204–12209.
 48. Konieczna P, Groeger D, Ziegler M, Frei R, Ferstl R, Shanahan F et al. Bifidobacterium infantis 35624 administration induces Foxp3 T regulatory cells in human peripheral blood: potential role for myeloid and plasmacytoid dendritic cells. *Gut* 2011; [Epub ahead of print].
 49. He SH, Chen X, Song CH, Liu ZQ, Zhou LF, Ma WJ et al. Interferon-lambda mediates oral tolerance and inhibits antigen-specific, T-helper 2 cell-mediated inflammation in mouse intestine. *Gastroenterology* 2011;**141**:249–258, 258 e241–242.
 50. Lai Y, Di Nardo A, Nakatsuji T, Leichterle A, Yang Y, Cogen AL et al. Commensal bacteria regulate Toll-like receptor 3-dependent inflammation after skin injury. *Nat Med* 2009;**15**:1377–1382.
 51. Lampen A, Meyer S, Arnhold T, Nau H. Metabolism of vitamin A and its active metabolite all-trans-retinoic acid in small intestinal enterocytes. *J Pharmacol Exp Ther* 2000;**295**:979–985.
 52. Rimoldi M, Chieppa M, Salucci V, Avogadri F, Sonzogni A, Sampietro GM et al. Intestinal immune homeostasis is regulated by the crosstalk between epithelial cells and dendritic cells. *Nat Immunol* 2005;**6**:507–514.
 53. Zaph C, Troy AE, Taylor BC, Berman-Booty LD, Guild KJ, Du Y et al. Epithelial-cell-intrinsic IKK-beta expression regulates intestinal immune homeostasis. *Nature* 2007;**446**:552–556.
 54. Ma D, Forsythe P, Bienenstock J. Live *Lactobacillus rhamnosus* [corrected] is essential for the inhibitory effect on tumor necrosis factor alpha-induced interleukin-8 expression. *Infect Immun* 2004;**72**:5308–5314.
 55. O'Hara AM, O'Regan P, Fanning A, O'Mahony C, Macsharry J, Lyons A et al. Functional modulation of human intestinal epithelial cell responses by *Bifidobacterium infantis* and *Lactobacillus salivarius*. *Immunology* 2006;**118**:202–215.
 56. Sibartie S, O'Hara AM, Ryan J, Fanning A, O'Mahony J, O'Neill S et al. Modulation of pathogen-induced CCL20 secretion from HT-29 human intestinal epithelial cells by commensal bacteria. *BMC Immunol* 2009;**10**:54.
 57. Kelly D, Campbell JI, King TP, Grant G, Jansson EA, Coutts AG et al. Commensal anaerobic gut bacteria attenuate inflammation by regulating nuclear-cytoplasmic shuttling of PPAR-gamma and RelA. *Nat Immunol* 2004;**5**:104–112.
 58. Neish AS, Gewirtz AT, Zeng H, Young AN, Hobert ME, Karmali V et al. Prokaryotic regulation of epithelial responses by inhibition of IkappaB-alpha ubiquitination. *Science* 2000;**289**:1560–1563.
 59. Di Giacinto C, Marinaro M, Sanchez M, Strober W, Boirivant M. Probiotics ameliorate recurrent Th1-mediated murine colitis by inducing IL-10 and IL-10-dependent TGF-beta-bearing regulatory cells. *J Immunol* 2005;**174**:3237–3246.
 60. O'Mahony C, Scully P, O'Mahony D, Murphy S, O'Brien F, Lyons A et al. Commensal-induced regulatory T cells mediate protection against pathogen-stimulated NF-kappaB activation. *PLoS Pathog* 2008;**4**: e1000112.
 61. Karim M, Kingsley CI, Bushell AR, Sawitzki BS, Wood KJ. Alloantigen-induced CD25⁺CD4⁺ regulatory T cells can develop in vivo from CD25⁺CD4⁺ precursors in a thymus-independent process. *J Immunol* 2004;**172**:923–928.
 62. Chen W, Jin W, Hardegen N, Lei KJ, Li L, Marinos N et al. Conversion of peripheral CD4⁺CD25⁺ naive T cells to CD4⁺CD25⁺ regulatory T cells by TGF-beta induction of transcription factor Foxp3. *J Exp Med* 2003;**198**:1875–1886.
 63. Coombes JL, Siddiqui KR, Arancibia-Carcamo CV, Hall J, Sun CM, Belkaid Y et al. A functionally specialized population of mucosal CD103⁺ DCs induces Foxp3⁺ regulatory T cells via a TGF-beta and retinoic acid-dependent mechanism. *J Exp Med* 2007;**204**:1757–1764.
 64. Sun CM, Hall JA, Blank RB, Bouladoux N, Oukka M, Mora JR et al. Small intestine lamina propria dendritic cells promote de novo generation of Foxp3 T reg cells via retinoic acid. *J Exp Med* 2007;**204**:1775–1785.
 65. Mazmanian SK, Liu CH, Tzianabos AO, Kasper DL. An immunomodulatory molecule of symbiotic bacteria directs maturation of the host immune system. *Cell* 2005;**122**:107–118.
 66. Russell DA, Ross RP, Fitzgerald GF, Stanton C. Metabolic activities and probiotic potential of bifidobacteria. *Int J Food Microbiol* 2011;**149**:88–105.
 67. Maslowski KM, Mackay CR. Diet, gut microbiota and immune responses. *Nat Immunol* 2011;**12**:5–9.
 68. Johansen H, Prescott SL. Practical prebiotics, probiotics and synbiotics for allergists: how useful are they? *Clin Exp Allergy* 2009;**39**:1801–1814.
 69. Schrezenmeier J, de Vrese M. Probiotics, prebiotics, and synbiotics—approaching a definition. *Am J Clin Nutr* 2001;**2**(Suppl):361S–364S.
 70. Cummings JH, Macfarlane GT. Gastrointestinal effects of prebiotics. *Br J Nutr* 2002;**87**(Suppl 2):S145–S151.
 71. Rao S, Srinivasjois R, Patole S. Prebiotic supplementation in full-term neonates: a systematic review of randomized controlled trials. *Arch Pediatr Adolesc Med* 2009;**163**:755–764.
 72. Sela DA, Chapman J, Adeyua A, Kim JH, Chen F, Whitehead TR et al. The genome sequence of *Bifidobacterium longum* subsp. *infantis* reveals adaptations for milk utilization within the infant microbiome. *Proc Natl Acad Sci U S A* 2008;**105**:18964–18969.
 73. Macfarlane S, Macfarlane GT. Regulation of short-chain fatty acid production. *Proc Nutr Soc* 2003;**62**:67–72.
 74. Hennessy AA, Ross RP, Devery R, Stanton C. Optimization of a reconstituted skim milk based medium for enhanced CLA production by bifidobacteria. *J Appl Microbiol* 2009;**106**:1315–1327.
 75. O'Shea EF, Cotter PD, Stanton C, Ross RP, Hill C. Production of bioactive substances by intestinal bacteria as a basis for explaining probiotic mechanisms: Bacteriocins and conjugated linoleic acid. *Int J Food Microbiol* 2012;**152**:189–205.
 76. Gobetti M, Cagno RD, De Angelis M. Functional microorganisms for functional food quality. *Crit Rev Food Sci Nutr* 2010;**50**:716–727.
 77. Karovicova J, Kohajdová Z. Biogenic Amines in Food. *Chem Pap* 2005;**59**:70–79.
 78. Muller I, Jenner J, Handgretinger R, Riberdy J, Kerst G. Glycosylation and lectins—examples of immunosurveillance and immune evasion. *Histol Histopathol* 2004;**19**:527–533.
 79. Drickamer K. Two distinct classes of carbohydrate-recognition domains in animal lectins. *J Biol Chem* 1988;**263**:9557–9560.
 80. van Kooyk Y, Rabinovich GA. Protein-glycan interactions in the control of innate and adaptive immune responses. *Nat Immunol* 2008;**9**:593–601.
 81. Rabinovich GA, Toscano MA. Turning 'sweet' on immunity: galectin-glycan interactions in immune tolerance and inflammation. *Nat Rev Immunol* 2009;**9**:338–352.
 82. Erbacher A, Gieseke F, Handgretinger R, Muller I. Dendritic cells: functional aspects of glycosylation and lectins. *Hum Immunol* 2009;**70**:308–312.
 83. Svajger U, Anderlüh M, Jeras M, Obermajer N. C-type lectin DC-SIGN: an

- adhesion, signalling and antigen-uptake molecule that guides dendritic cells in immunity. *Cell Signal* 2010;**22**:1397–1405.
84. van Kooyk Y. C-type lectins on dendritic cells: key modulators for the induction of immune responses. *Biochem Soc Trans* 2008;**6**:1478–1481.
 85. Hawiger D, Inaba K, Dorsett Y, Guo M, Mahnke K, Rivera M et al. Dendritic cells induce peripheral T cell unresponsiveness under steady state conditions in vivo. *J Exp Med* 2001;**194**:769–779.
 86. van Kooyk Y, Geijtenbeek TB. DC-SIGN: escape mechanism for pathogens. *Nat Rev Immunol* 2003;**3**:697–709.
 87. Gringhuis SI, den Dunnen J, Litjens M, van Het Hof B, van Kooyk Y, Geijtenbeek TB. C-type lectin DC-SIGN modulates Toll-like receptor signaling via Raf-1 kinase-dependent acetylation of transcription factor NF-kappaB. *Immunity* 2007;**26**:605–616.
 88. Zhou Y, Kawasaki H, Hsu SC, Lee RT, Yao X, Plunkett B et al. Oral tolerance to food-induced systemic anaphylaxis mediated by the C-type lectin SIGNR1. *Nat Med* 2010;**16**:1128–1133.
 89. Royer PJ, Emara M, Yang C, Al-Ghoulh A, Tighe P, Jones N et al. The mannose receptor mediates the uptake of diverse native allergens by dendritic cells and determines allergen-induced T cell polarization through modulation of IDO activity. *J Immunol* 2010;**185**:1522–1531.
 90. Cerliani JP, Stowell SR, Mascanfroni ID, Arthur CM, Cummings RD, Rabinovich GA. Expanding the universe of cytokines and pattern recognition receptors: galectins and glycans in innate immunity. *J Clin Immunol* 2011;**31**:10–21.
 91. de Kivit S, Kraneveld AD, Garssen J, Willemssen LE. Glycan recognition at the interface of the intestinal immune system: target for immune modulation via dietary components. *Eur J Pharmacol* 2011;**668**:124–132.
 92. Chen HY, Sharma BB, Yu L, Zuberi R, Weng IC, Kawakami Y et al. Role of galectin-3 in mast cell functions: galectin-3-deficient mast cells exhibit impaired mediator release and defective JNK expression. *J Immunol* 2006;**177**:4991–4997.
 93. Crocker PR. Siglecs in innate immunity. *Curr Opin Pharmacol* 2005;**5**:431–437.
 94. Crocker PR, Paulson JC, Varki A. Siglecs and their roles in the immune system. *Nat Rev Immunol* 2007;**7**:255–266.
 95. Crocker PR, Redelinghuys P. Siglecs as positive and negative regulators of the immune system. *Biochem Soc Trans* 2008;**36** (Pt 6):1467–1471.
 96. Jandus C, Simon HU, von Gunten S. Targeting siglecs—a novel pharmacological strategy for immuno- and glycotherapy. *Biochem Pharmacol* 2011;**82**:323–332.
 97. McMillan SJ, Crocker PR. CD33-related sialic-acid-binding immunoglobulin-like lectins in health and disease. *Carbohydr Res* 2008;**343**:2050–2056.
 98. Ando M, Tu W, Nishijima K, Iijima S. Siglec-9 enhances IL-10 production in macrophages via tyrosine-based motifs. *Biochem Biophys Res Commun* 2008;**369**:878–883.
 99. von Gunten S, Bochner BS. Basic and clinical immunology of Siglecs. *Ann N Y Acad Sci* 2008;**1143**:61–82.
 100. Nutku E, Aizawa H, Hudson SA, Bochner BS. Ligation of Siglec-8: a selective mechanism for induction of human eosinophil apoptosis. *Blood* 2003;**101**:5014–5020.
 101. Bochner BS. Siglec-8 on human eosinophils and mast cells, and Siglec-F on murine eosinophils, are functionally related inhibitory receptors. *Clin Exp Allergy* 2009;**39**:317–324.
 102. Yokoi H, Choi OH, Hubbard W, Lee HS, Canning BJ, Lee HH et al. Inhibition of FcεpsilonRI-dependent mediator release and calcium flux from human mast cells by sialic acid-binding immunoglobulin-like lectin 8 engagement. *J Allergy Clin Immunol* 2008;**121**:499.e1–505.e1.
 103. Meijer K, de Vos P, Priebe MG. Butyrate and other short-chain fatty acids as modulators of immunity: what relevance for health? *Curr Opin Clin Nutr Metab Care* 2010;**13**:715–721.
 104. Maslowski KM, Vieira AT, Ng A, Kranich J, Sierro F, Yu D et al. Regulation of inflammatory responses by gut microbiota and chemoattractant receptor GPR43. *Nature* 2009;**461**:1282–1286.
 105. Brown AJ, Goldsworthy SM, Barnes AA, Eilert MM, Tcheang L, Daniels D et al. The Orphan G protein-coupled receptors GPR41 and GPR43 are activated by propionate and other short chain carboxylic acids. *J Biol Chem* 2003;**278**:11312–11319.
 106. Le Poul E, Loison C, Struyf S, Springael JY, Lannoy V, Decobecq ME et al. Functional characterization of human receptors for short chain fatty acids and their role in polymorphonuclear cell activation. *J Biol Chem* 2003;**278**:25481–25489.
 107. Yonezawa T, Kobayashi Y, Obara Y. Short-chain fatty acids induce acute phosphorylation of the p38 mitogen-activated protein kinase/heat shock protein 27 pathway via GPR43 in the MCF-7 human breast cancer cell line. *Cell Signal* 2007;**19**:185–193.
 108. Vinolo MA, Ferguson GJ, Kulkarni S, Damoulakis G, Anderson K, Bohlooly YM et al. SCFAs induce mouse neutrophil chemotaxis through the GPR43 receptor. *PLoS ONE* 2011;**6**:e21205.
 109. Dianzani C, Cavalli R, Zara GP, Gallicchio M, Lombardi G, Gasco MR et al. Cholesteryl butyrate solid lipid nanoparticles inhibit adhesion of human neutrophils to endothelial cells. *Br J Pharmacol* 2006;**148**:648–656.
 110. Cox MA, Jackson J, Stanton M, Rojas-Triana A, Bober L, Lavery M et al. Short-chain fatty acids act as antiinflammatory mediators by regulating prostaglandin E(2) and cytokines. *World J Gastroenterol* 2009;**15**:5549–5557.
 111. Singh N, Thangaraju M, Prasad PD, Martin PM, Lambert NA, Boettger T et al. Blockade of dendritic cell development by bacterial fermentation products butyrate and propionate through a transporter (Slc5a8)-dependent inhibition of histone deacetylases. *J Biol Chem* 2010;**285**:27601–27608.
 112. Spiegelberg BD, Hamm HE. G betagamma binds histone deacetylase 5 (HDAC5) and inhibits its transcriptional co-repression activity. *J Biol Chem* 2005;**280**:41769–41776.
 113. Diakos C, Prieschl EE, Saemann MD, Bohmig GA, Csonga R, Sobanov Y et al. n-Butyrate inhibits Jun NH(2)-terminal kinase activation and cytokine transcription in mast cells. *Biochem Biophys Res Commun* 2006;**349**:863–868.
 114. Assem el SK, Peh KH, Wan BY, Middleton BJ, Dines J, Marson CM. Effects of a selection of histone deacetylase inhibitors on mast cell activation and airway and colonic smooth muscle contraction. *Int Immunopharmacol* 2008;**8**:1793–1801.
 115. Talukdar S, Olefsky JM, Osborn O. Targeting GPR120 and other fatty acid-sensing GPCRs ameliorates insulin resistance and inflammatory diseases. *Trends Pharmacol Sci* 2011;**32**:543–550.
 116. Oh DY, Talukdar S, Bae EJ, Imamura T, Morinaga H, Fan W et al. GPR120 is an omega-3 fatty acid receptor mediating potent anti-inflammatory and insulin-sensitizing effects. *Cell* 2010;**142**:687–698.
 117. Zapata-Gonzalez F, Rueda F, Petriz J, Domingo P, Villarroya F, Diaz-Delfin J et al. Human dendritic cell activities are modulated by the omega-3 fatty acid, docosahexaenoic acid, mainly through PPAR (gamma):RXR heterodimers: comparison with other polyunsaturated fatty acids. *J Leukoc Biol* 2008;**84**:1172–1182.
 118. Straus DS, Glass CK. Anti-inflammatory actions of PPAR ligands: new insights on cellular and molecular mechanisms. *Trends Immunol* 2007;**28**:551–558.
 119. Brix S, Lund P, Kjaer TM, Straarup EM, Hellgren LI, Frokiaer H. CD4(+) T-cell activation is differentially modulated by bacteria-primed dendritic cells, but is

- generally down-regulated by n-3 polyunsaturated fatty acids. *Immunology* 2010;**129**:338–350.
120. Calder PC, Kremmyda LS, Vlachava M, Noakes PS, Miles EA. Is there a role for fatty acids in early life programming of the immune system? *Proc Nutr Soc* 2010;**69**:373–380.
121. O'Shea M, Bassaganya-Riera J, Mohede IC. Immunomodulatory properties of conjugated linoleic acid. *Am J Clin Nutr* 2004;**6** (Suppl):1199S–1206S.
122. Beghdadi W, Porcherie A, Schneider BS, Dubayle D, Peronet R, Huerre M et al. Inhibition of histamine-mediated signaling confers significant protection against severe malaria in mouse models of disease. *J Exp Med* 2008;**205**:395–408.
123. O'Mahony L, Akdis M, Akdis CA. Regulation of the immune response and inflammation by histamine and histamine receptors. *J Allergy Clin Immunol* 2011;**128**: 1153–1162.
124. Siragusa S, De Angelis M, Di Cagno R, Rizzello CG, Coda R, Gobbetti M. Synthesis of gamma-aminobutyric acid by lactic acid bacteria isolated from a variety of Italian cheeses. *Appl Environ Microbiol* 2007;**73**:7283–7290.
125. Xiang YY, Wang S, Liu M, Hirota JA, Li J, Ju W et al. A GABAergic system in airway epithelium is essential for mucus overproduction in asthma. *Nat Med* 2007;**13**:862–867.