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REVIEW

Gut microbiota in regulatory T cell generation and function: mechanisms and health implications

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ABSTRACT

The establishment and maintenance of immune homeostasis rely on a dynamic, bidirectional exchange of information between commensal microorganisms and the host immune system. At the center of this process are $CD4^+Foxp3^+$ regulatory T cells (Tregs), which have emerged as pivotal mediators to ensure immunological equilibrium. This review explores the sophisticated mechanisms by which the gut microbiota modulates the differentiation, expansion, and functional specialization of Tregs, orchestrating intestinal immune tolerance to support host-microbiota mutualism. We discuss the role of microbial-derived structural components and metabolites in shaping the immunoregulatory fitness of Tregs. Additionally, we explore the impact of gut microbial dysbiosis, where disrupted microbial-immune crosstalk compromises immune tolerance, contributing to the development of inflammatory and autoimmune disorders. Finally, we highlight the potential of microbiota-based strategies to recalibrate intestinal immunity and restore immune tolerance.

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Introduction

The human body represents a complex ecosystem, intricately intertwined with trillions of microorganisms that collectively constitute the gut microbiome – a dynamic consortium encompassing bacteria, viruses, fungi, and other microbial entities. This microbial community is dominated by two bacterial phyla, *Bacteroidetes* and *Firmicutes*, which constitute approximately 90% of the total gut microbial biomass, with smaller contributions from *Proteobacteria*, *Actinobacteria*, and *Verrucomicrobia*.[1–3] Commensal fungi, such as *Candida* and *Saccharomyces* species, and *bacteriophages* further contribute to this ecosystem, while gut-resident bacteria reciprocally suppress pathogenic invaders. While the human genome encodes approximately 20,000 genes, the hologenome, which integrates the host genome with the collective genetic material of its resident microbiota, comprises over 33 million genes.[4] This vast genetic reservoir enables the gut microbiota to establish a mutualistic relationship with the host, performing critical functions such as fermenting dietary fibers to produce short-chain fatty acids (SCFAs),[5–7] synthesizing vitamins (e.g., vitamin B12 and K),[8,9] metabolizing xenobiotics,[10] and competitively excluding pathogens,[11] while the human gut provides protection, nutrients, and favorable growth conditions for these microbes.

Additionally, the gut microbiome has emerged as a central regulator of host immunity, profoundly influencing immune development, tolerance, and homeostasis influencing both innate and adaptive immune responses.[5,12] Dysbiosis – alterations in microbial composition or function – has been linked to numerous diseases, including inflammatory bowel disease (IBD), autoimmune disorders, and cancer.[5,12–16]

For this mutualistic relationship to thrive, the host needs to recognize the microbiome as part of itself, a process facilitated by co-evolved

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mechanisms that ensure immune tolerance. The gut immune system comprises a highly specialized and compartmentalized network of innate and adaptive immune components that work synergistically to maintain mucosal homeostasis and defend against pathogens. Key innate immune cells involved in mucosal defense include intestinal epithelial cells (IECs) and innate lymphoid cells (ILCs). IECs act as a physical barrier and also produce antimicrobial peptides and cytokines. Among the ILCs, group 3 ILCs (ILC3s) are especially important for maintaining mucosal immune homeostasis and promoting tolerance to commensal microbes.[17,18] Dendritic cells (DCs) and macrophages continuously sample luminal antigens and help orchestrate immune responses, often promoting regulatory over inflammatory pathways.[19,20] Among adaptive components, IgA-producing plasma cells are essential for neutralizing pathogens and shaping microbial composition without inducing inflammation.[21] Importantly, the gut harbors a substantial population of type 1 regulatory (Tr1) T cells that secrete high levels of IL-10 and do not express Foxp3 constitutively.[22] Additionally, regulatory B cells that produce IL-10 and TGF-β to suppress inflammatory responses.[23] $Foxp3^{+}CD4^{+}$ regulatory T cells (Tregs) and Th17 cells, whose balance is crucial for immune tolerance and pathogen defense, respectively.[12]

Tregs, a specialized subset of $CD4^{+}$ T cells characterized by the expression of the transcription factor Foxp3, play a crucial role in maintaining immune homeostasis and preventing excessive inflammatory responses.[24,25] Tregs are indispensable for establishing dominant immune tolerance and maintaining immune homeostasis. Tregs exert their suppressive functions through multiple mechanisms, including the production of anti-inflammatory cytokines (IL-10, TGF-β, IL-35), metabolic disruption of effector T cells, cytolysis, and modulation of dendritic cell function.[24,25] They are broadly classified into two categories: thymic Tregs (tTregs), which develop in the thymus and prevent autoimmunity, and peripheral Tregs (pTregs), which differentiate in peripheral tissues and mediate tolerance to innocuous antigens, including dietary components and commensal microbes.[15,26] Within the intestinal mucosa, a significant population of Tregs co-expresses Foxp3 and RORγt, the latter being a transcription factor typically associated with Th17 cells.[15] These RORγt + Tregs are predominantly of peripheral origin and play a crucial role in maintaining tolerance to the gut microbiota.[26] Their development and maintenance are heavily influenced by microbial signals, highlighting the intimate relationship between the gut microbiota and the regulatory arm of the immune system.

The gut microbiome has evolved sophisticated mechanisms to influence the differentiation, expansion, and functional fitness of Tregs. In turn, Tregs suppress excessive immune responses, thereby preserving the diversity and eubiosis of the commensal microbiota. This reciprocal interaction underscores the critical importance of the microbiome-Treg axis in immune regulation. Recent advances have revealed that microbial structural components, such as polysaccharide A (PSA), cell surface β-glucan/galactan polysaccharides (CSGG), and mannan/β-1,6-glucan-containing polysaccharides (MGCP), directly modulate Treg differentiation and function.[27–29] Furthermore, microbial metabolites – including short-chain fatty acids (SCFAs), tryptophan derivatives, and BA – play pivotal roles in shaping Treg biology through epigenetic modifications, metabolic reprogramming, and receptor-mediated signaling pathways.[30–32] However, dysregulation of the microbiome-Treg axis can lead to immune dysfunction, contributing to the pathogenesis of inflammatory and autoimmune diseases. For example, in inflammatory bowel disease (IBD), dysbiosis and reduced production of SCFAs and secondary BA impair Treg function, resulting in chronic inflammation.[13,33]

In this review, we explore microbial factors and mechanisms that support Treg function in maintaining immune homeostasis. We also examine the therapeutic potential of targeting the microbiome-Treg axis in the context of inflammatory and autoimmune diseases. By integrating recent advances, we highlight the pivotal role of microbially derived signals in immune regulation and their implications for disease prevention and treatment.

Microbial modulation of tregs

The incorporation of gut microbes into the host's immunological self requires the establishment of

active immune tolerance to prevent inappropriate immune activation while preserving the ability to respond to harmful pathogens. Indeed, Germ-free (GF) mice, which lack a gut microbiome, exhibit an underdeveloped immune system, highlighting the critical role of microbial colonization in immune maturation.[5] Furthermore, the depletion of microbiota with oral antibiotics has been shown to exacerbate intestinal inflammation, underscoring the importance of the gut microbiome in maintaining peripheral tolerance.[34] Tregs have been extensively studied toward establishment of central and peripheral immune tolerance, since their discovery and, over the last two decades, have emerged as central regulators in establishing and maintaining dominant immune tolerance.[35] As described above, two major subtypes of Tregs – tTregs and pTregs were initially thought to have distinct roles, recent studies suggest that both subsets can be induced in response to microbial antigens, challenging their traditional classifications.[36,37]

The gut microbiota plays a critical role in shaping Treg populations, both in the thymus and the periphery. Microbial-derived signals, including polysaccharides, metabolites, and structural components, directly influence Treg differentiation, expansion, and function.[5,31] These interactions highlight the intricate crosstalk between the microbiota and Tregs, which is essential for maintaining immune homeostasis and preventing inflammatory diseases.

While the gut microbiota is a critical regulator of Treg-mediated immune tolerance, not all microbial species or their metabolites universally promote anti-inflammatory responses. Certain gut bacteria, such as Segmented Filamentous Bacteria (SFB), are known to drive pro-inflammatory Th17 cell responses, which can exacerbate inflammation in susceptible hosts.[34,38] For instance, SFB colonization in mice has been shown to promote Th17 cell differentiation in the gut, contributing to autoimmune conditions such as experimental autoimmune encephalomyelitis (EAE).[39] Similarly, *Prevotella copri* has been associated with enhanced susceptibility to colitis and arthritis through activation of pro-inflammatory pathways.[40,41] Pathobionts such as *Enterococcus faecalis* and adherent-invasive *Escherichia coli* (AIEC) exacerbate inflammatory bowel disease (IBD) by activating NF-κB and NLRP3 inflammasome pathways, thereby suppressing Treg activity.[42–44] Even commensals like Helicobacter hepaticus can adopt pathogenic roles in genetically susceptible hosts, triggering colitis through IL-23-driven Th17 responses.[45] These examples underscore the context-dependent nature of microbial-immune interactions, where the same microbiota can either promote tolerance or inflammation depending on host genetics, microbial strain specificity, and environmental triggers. In this section, we explore the mechanisms by which the relevant gut microbiota modulates Treg biology, focusing on the generation and function of both thymic and pTregs in response to microbial components.

Microbial regulation of thymic treg development

GF mice generally display a reduced thymus size, indicating the importance of microbiota in thymic cellular development and immune maturation.[5] Within the thymus, medullary thymic epithelial cells (mTECs) play a central role in establishing central self-tolerance. They achieve this through the negative selection of self-reactive T cells via clonal deletion or their differentiation into Tregs. This process is facilitated by the promiscuous expression of tissue-restricted antigens (TRAs), driven by transcription factors such as Aire[46,47] and Fezf2.[48] Intriguingly, mTECs also express multiple Toll-like receptors (TLRs), suggesting a potential role for microbial signals in thymic Treg development.[49] While TLR signaling has been shown to be important for Treg generation, there is no significant difference in TLR-MyD88-mediated cytokine gene expression between mTECs from GF and specific pathogen-free (SPF) mice. This indicates that mTEC TLRs may be activated by endogenous ligands rather than microbial signals.[49]

Both mTEC and thymic DCs can present antigens to drive Treg cell generation.[50,51] During a critical period of early neonatal life in mice, intestinal CX3CR1^{+} dendritic cells transport microbial antigens from the intestine to the thymus. Interestingly, these antigens primarily stimulate microbiota-specific conventional T cells rather than tTregs.[52] However, this study utilized Segmented filamentous bacteria (SFB) as a model microorganism, predominantly inducing Th17 T

cell responses, thus it remains to be seen if Treg-inducing bacteria could expand tTregs under similar settings.

Further evidence of microbial influence on tTregs comes from studies using limited T cell receptor (TCR) models. The TCR repertoire of tTregs was found to be significantly overlapping with colonic Tregs,[37] suggesting shared antigen specificity between these populations. In mice deficient in extra-thymic Treg generation, a niche of tTregs is established in early post-natal life. Interestingly, these cells proliferate independent of IL-2 signaling but require microbial antigens for their expansion, highlighting the role of microbial signals in shaping thymic Treg dynamics.[36]

Despite these insights, the precise contribution of microbial signals to thymic Treg development remains unclear. The lack of definitive markers and the interchangeability between Treg subsets make it challenging to unequivocally determine the thymic origin of microbiota-induced Tregs. Future studies employing lineage-tracing models and single-cell technologies, as well as monocolonization studies with Treg-inducing bacteria, will be essential to dissect the mechanisms by which microbial signals influence thymic Treg development and function.

Microbial antigen-induced pTregs

Immune tolerance to gut microbiota is primarily mediated by peripheral RORγt$^+$ Tregs (RORγt$^+$ pTregs), which arise from naïve conventional CD4$^+$ T cells under specific activation conditions.[6,7] These RORγt$^+$ pTregs populate the gut mucosal immune system during a critical developmental window around weaning in mice, coinciding with robust microbial colonization of the gut.[28] The generation of these pTregs depends on bacterial antigens, diet-derived metabolites, and host-produced retinoic acid.[28] Interestingly, disruptions to the microbiota during this early life period can lead to inflammatory pathologies later in life, underscoring the importance of this temporal window in establishing immune tolerance.[28]

Gut microbiota is essential for generating pTreg diversity and their functional fitness in the colon.[53] Despite significant progress in understanding pTreg biology, the identity of the antigen-presenting cells (APCs) responsible for mediating their induction has remained elusive. CD103$^+$ conventional dendritic cells (cDCs) have been implicated in promoting pTreg differentiation in response to luminal antigens.[20,54–59] However, studies using adoptive transfer models have demonstrated that *Helicobacter*-specific T cells can differentiate into pTregs even in the absence of CD103$^+$ DCs, suggesting that these cells are not indispensable for microbial antigen-driven pTreg generation.[60]

Recent investigations have highlighted the potential role of RORγt-expressing APCs in pTreg induction[61–63] (Figure 1). Consistent with previous observations,[60] these studies excluded a role for conventional DCs in this process. Deletion of MHCII from RORγt$^+$ APCs resulted in a marked reduction in gut RORγt$^+$ pTregs. Lyu et al. identified lymphoid tissue inducer (LTi)-like group3 ILC (ILC3) as key players in RORγt$^+$ pTreg generation through antigen presentation and integrin αvβ3-mediated processing of latent TGFβ.[63] Notably, this study provided the first evidence implicating integrin αvβ3 in pTreg induction. In a mouse model where MHCII was deleted specifically in ILC3s (*H2-Ab1*$^{fl/fl}$*Rorc*^{Cre}), a significant reduction in RORγt$^+$ pTregs was observed in mLNs and large intestine. Furthermore, a correlation between ILC3s and RORγt$^+$ pTregs was observed in the human intestine, with a disruption of these cells noted in patients with IBD.[63] Kedmi et al. demonstrated that RORγt$^+$ APCs (which were either ILC3 or Janus type cells) require CCR7-mediated migration, MHCII-dependent antigen presentation, and integrin αvβ8 functionality to effectively induce RORγt$^+$ pTregs.[62] When these processes are impaired, the failure to generate pTregs results in the expansion of pathogenic Th17 cells instead.

In contrast, Akagbosu et al. identified Thetis cells, a distinct subset of RORγt$^+$ APCs, as key mediators of pTreg generation during early life. Specifically, subgroup TC IV, characterized by the expression of *Itgav* and *Itgb8* (encode Integrin subunit α_v and β_8, respectively) and *Tgfb2* (encodes TGFβ), was shown to play an essential role in this process.[61] In contrast to the findings reported by Lyu et al., Akagbosu et al. demonstrated that ILC3s were dispensable for RORγt$^+$ pTreg generation using *H2-Ab1*$^{fl/fl}$*Rora*^{Cre} mice, which selectively

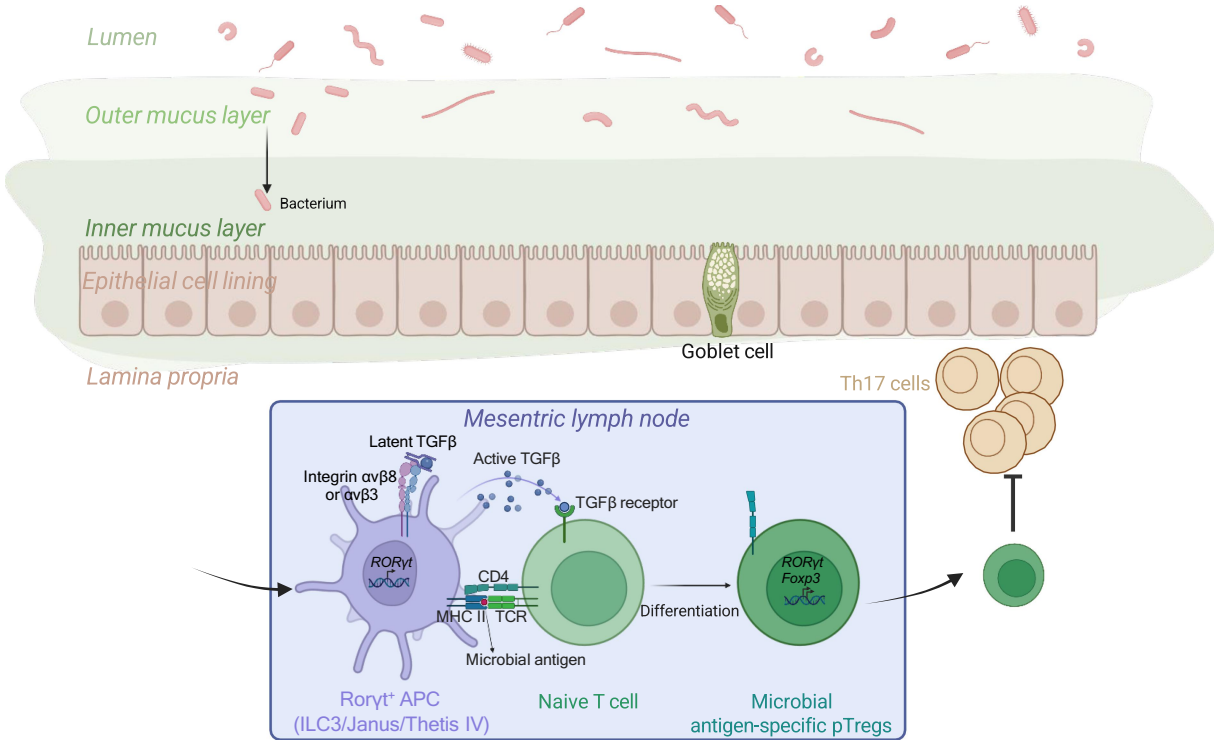

Figure 1. Mechanism of microbial antigen-specific pTreg generation in mesenteric lymph nodes. In the gut, bacterial antigens are captured by RORγt^{+} antigen-presenting cells (APCs) that express either integrin αvβ8 or αvβ3, which activate latent TGF-β. These APCs present microbial antigens to naïve T cells in an MHC class II-restricted manner while simultaneously activating latent TGF-β. This process drives the differentiation of naïve CD4^{+} T cells into CD4^{+}RORγt^{+}Foxp3^{+} peripheral regulatory T cells (pTregs) in the presence of active TGF-β.

delete MHCII in ILC3. This discrepancy may arise from differences in Cre drivers used in the respective mouse models, suggesting the possibility that *Rorc*^{Cre}-mediated deletion might also affect MHCII expression in Thetis cells. Resolving this issue will require the development of genetic tools to specifically target Thetis cells. Further, by analyzing the single-cell atlas of human intestinal and gut-draining lymph node cells spanning fetal to adult life,[64] Akagbosu et al. identified a cluster within the myeloid cells that expressed signature Thetis cell genes – *TNFRSF11B* and *SPIB*, along with *AIRE*. These cells were predominantly localized in mLNs and enriched in fetal samples, suggesting a potential role in establishing gut immune tolerance early in life. Whether these cells functionally contribute to the establishment of tolerance to gut microbiota in humans remains to be determined. Future studies are needed to elucidate their mechanistic roles and validate their functional significance in peripheral immune tolerance.

The human leukocyte antigen (HLA) system plays a crucial role in shaping the interaction between microbial antigens and the host immune system, including the development of Treg cells. Certain HLA alleles have been associated with altered susceptibility to autoimmune and inflammatory conditions, which may be partly mediated through their influence on microbiota-Treg interactions.[65] For instance, HLA-DQ2 and HLA-DQ8 haplotypes, which are strongly associated with celiac disease (CeD), influence the presentation of both gluten peptides and potentially microbial antigens that may share structural similarities.[66] This molecular mimicry could affect Treg induction and function in genetically susceptible individuals. Furthermore, recent studies have demonstrated that specific HLA alleles can influence the composition of the gut microbiota,[67] potentially creating a feedback loop that affects Treg homeostasis. The HLA-microbiota-Treg axis represents an important area for future research, particularly in understanding how genetic factors influence individual responses to microbial antigens and subsequent immune regulation.

Microbe-derived ligands in treg generation

Bacterial structural components, such as lipopolysaccharides, peptidoglycans etc. interact with diverse host immune receptors, including TLRs and NOD-like receptors (NLRs), to shape the immune landscape. While the adjuvant effect of microbial components in activating the effector immune response is well established, we and others have demonstrated their equally critical role in driving immunoregulatory responses.[27,29,68,69] Our previous work has demonstrated that a probiotic mixture named IRT5, comprising *Lactobacillus acidophilus*, *Lactobacillus casei*, *Lactobacillus reuteri*, *Bifidobacterium bifidum*, and *Streptococcus thermophilus*, induces the generation of Foxp3$^+$ Tregs.[70] This process is mediated by tolerogenic DCs that express high levels of IL-10, TGF-β, COX-2, and indoleamine 2,3-dioxygenase (IDO). Similarly, *Lactobacillus pentosus* KF340 (LP340) induced IL-10 Type 1 regulatory T cells (Tr1 cells), alleviating atopic dermatitis in mice.[71] However, the specific effector components responsible for these immunomodulatory effects remained unidentified. Identifying these effector components is crucial for comprehending the molecular language of host-microbiome interactions. Moreover, this knowledge is essential for developing prebiotics, probiotics, and live biotherapeutic products (LBP) with a broad therapeutic window. To address this gap, we recently have rationally identified a unique dietary commensal strain, *Lactiplantibacillus plantarum* IMB19 (LpIMB19), and its effector component capsular rhamnose-rich heteropolysaccharide (RHP), which has the capability to enhance CD8 T cell immune response and augment anti-tumor immunity.[72,73] The RHP functions as a TLR2 ligand, modulating tumor-associated macrophages toward an inflammatory phenotype, which subsequently activates CD8 T cells. To modulate Treg-mediated immunoregulatory responses, we and other researchers have identified specific microbial ligands capable of enhancing both the frequency and suppressive function of Tregs. These ligands have been shown to effectively alleviate disease progression in various mouse models of gut-related disorders as well as pathologies affecting distant tissues.

Polysaccharide a (PSA)

In a significant study, Mazmanian et al.[28] identified PSA, a protease-resistant zwitterionic capsular polysaccharide derived from the human commensal bacterium *Bacteroides fragilis*, as the first example of a unique symbiont molecule capable of promoting immunoregulatory responses. PSA was shown to directly interact with TLR2 on T cells, driving the induction and expansion of Tregs and, thus, suppressing the differentiation of pro-inflammatory Th17 cells[74] (Figure 2). This discovery established a foundational framework for the rational identification of commensal bacteria with Treg-inducing properties, offering new avenues for modulating immune tolerance. However, subsequent studies revealed additional layers of complexity in PSA-mediated immunomodulation. In an in vitro co-culture system, Kreisman et al.[75] demonstrated that human CD4$^+$ T cells exposed to PSA in the presence of a mixed population of APCs differentiated into IL-10-producing Tr1 cells, which are distinct from Foxp3$^+$ Tregs. Notably, Telesford et al.[76] found that the ability of PSA to induce Foxp3$^+$ Tregs was dependent on DCs, suggesting that DC-mediated processing and presentation of PSA are critical for its Treg-inducing effects. This finding underscores the critical role of DC-mediated processing and presentation of PSA in shaping its Treg-inducing effects and highlights how specific APC subsets influence the nature of the T cell response elicited by PSA. The clinical relevance of PSA-producing *B. fragilis* has been further emphasized by studies showing a reduced prevalence of actively PSA-producing strains in colonic biopsies from patients with IBD.[77,78] These observations suggest that the loss of PSA-mediated immunoregulatory signals may contribute to the dysregulated immune responses characteristic of IBD, underscoring the therapeutic potential of PSA and PSA-producing bacteria in restoring immune homeostasis. Genomic screening has identified various commensal bacteria, including some pathogens, that produce capsular zwitterionic polysaccharides akin to PSA. Notably, *Bacteroides cellulosilyticus* DSM 14,838 was shown to protect against colitis in mice,[79] underscoring zwitterionic polysaccharides as a promising class of immunomodulatory molecules for therapeutic use.

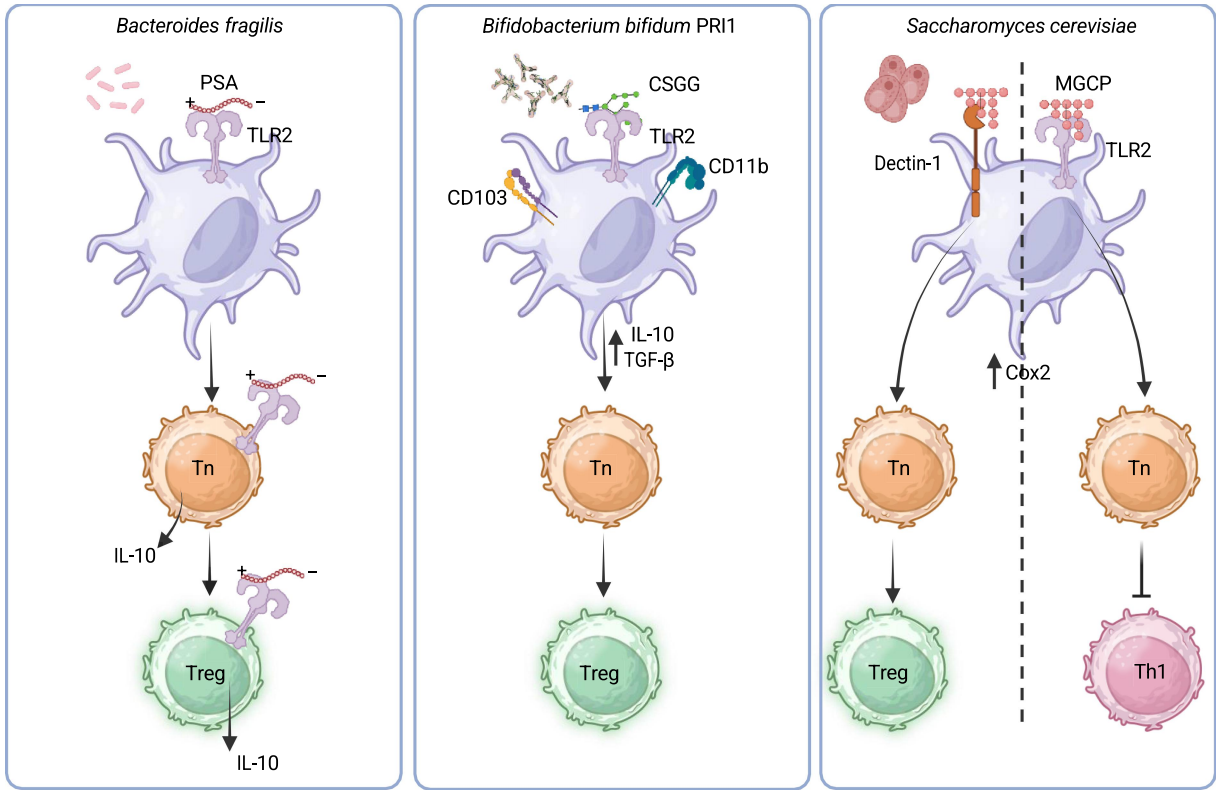

Figure 2. Microbial ligands drive pTreg generation and modulate immune responses. Left Panel: *Bacteroides fragilis* produces a protease-resistant zwitterionic capsular polysaccharide known as Polysaccharide A (PSA), which acts as a ligand for Toll-like receptor 2 (TLR2). Upon binding to TLR2, PSA induces dendritic cells (DCs) to adopt a regulatory phenotype, promoting the differentiation of naïve CD4+ T cells (Tn) into peripheral regulatory T cells (pTregs). Additionally, PSA can directly interact with TLR2 on both naïve T cells and Tregs, stimulating the production of the anti-inflammatory cytokine IL-10 Middle Panel: *Bifidobacterium bifidum* strain PRI1 (Bb PRI1) expresses cell surface β-glucan/galactan polysaccharides (CSGG), which are potent inducers of pTregs. CSGG binds to TLR2 on $CD103^{+}CD11b^{+}$ DCs, driving these cells toward a tolerogenic phenotype characterized by the production of IL-10 and TGF-β. This environment promotes the differentiation of naïve $CD4^{+}$ T cells into $CD4^{+}Foxp3^{+}$ pTregs. Right Panel: Polysaccharides derived from commensal yeast cell walls, such as mannan/β-1,6-glucan-containing polysaccharides (MGCP), are strong inducers of pTregs and inhibit the differentiation of inflammatory Th1 cells. MGCP operates through two distinct pathways in DCs: 1. Binding to Dectin-1 on DCs enhances Cox2 production, fostering a tolerogenic phenotype that supports the differentiation of naïve $CD4^{+}$ T cells into $CD4^{+}Foxp3^{+}$ pTregs. 2. MGCP-treated DCs suppress Th1 cell differentiation and IFN-γ production in a Cox2-dependent manner. Notably, this suppression requires MGCP binding to TLR2 on DCs.

Cell surface - β glucan/galactan polysaccharides (CSGG)

Through extensive ex vivo screening to identify bacteria capable of inducing pTregs, we discovered that *Bifidobacterium bifidum* strain PRI1 (*Bb* PRI1) possesses significant pTreg-inducing properties.[29] *Bifidobacterium* species are well-documented for their ability to colonize the gut of breastfed infants early in life, playing a critical role in shaping the neonatal immune system.[80] Notably, supplementation with *Bifidobacterium* has been shown to alleviate allergic inflammation in infants with dysbiotic gut microbiota compositions.[81] In GF mice mono-colonized with Bb PRI1, the strain was found to promote the development of $CD103^{+}$ $CD11b^{+}$ regulatory DCs in the colon. Intriguingly, monocolonization of GF mice with Bb PRI1 induced colonic Tregs with relatively diverse TCR clonotypes. These pTregs were not only reactive to the bacterium itself but also expanded in response to dietary antigen OVA and bacterial flagellin. To further explore how Bb PRI1 influences the functional orientation of colonic Treg cells with distinct TCR repertoires, we conducted single-cell RNA sequencing and performed a comparative analysis of colonic Tregs from both SPF and GF mice.[53] Our findings indicate that Bb PRI1 could alter the activation trajectory of colonic

Tregs, promoting the emergence of a distinct phenotypic subset that is prevalent in SPF mice but absent in GF mice. Additionally, Bb PRI1 exposure facilitated the expansion of specific Treg clonotypes characterized by shared transcriptional features. The microbiota-driven colonic Treg subset, identified as PD-1− CXCR3$^+$ Tregs, exhibited greater suppressive capacity than their counterparts from GF mice, demonstrated increased IL-10 production, and played a central role in modulating enteric inflammation in dextran sodium sulfate (DSS)-induced colitis.[53]

Fractionation of Bb PRI1's cellular components revealed that its cell surface CSGG were critical mediators of Treg induction. CSGG is a complex mixture of neutral polysaccharides, including β-1–6-glucan, β-1–4-galactan, β-1–6-galactan, and β-galactofuranan, which collectively act as ligands for TLR2.[82] Engagement of TLR2 by CSGG triggers DCs to produce IL-10 and TGF-β. CSGG acts as a ligand for TLR2, triggering DCs to produce the anti-inflammatory cytokines IL-10 and TGF-β, fostering an immunoregulatory environment (Figure 2). While CSGG's ability to activate TLR2 and induce DC-mediated production of IL-10 and TGF-β has been established, further research is needed to elucidate the downstream signaling pathways activated by TLR2 engagement and their precise role in mediating these immunomodulatory effects. Importantly, CD4$^+$ Foxp3$^+$ Tregs induced by CSGG treatment demonstrated functional activity, effectively suppressing the progression of inflammatory colitis in mouse models. It is to be noted that in Tregs, TLR signaling can have context-dependent effects. TLR2 activation by certain bacterial lipopeptides can temporarily abrogate the suppressive function of Tregs by inducing a shift toward a Th17-like phenotype, characterized by reduced Foxp3 expression and increased IL-17 production.[83] This effect is mediated through the MyD88-dependent activation of NF-κB and PI3K/Akt pathways, which inhibit Foxp3 function.[84] Conversely, TLR2 signaling can also promote Treg expansion under certain conditions like CSGG treatment, highlighting the context-dependent nature of these pathways.[29,74]

Mannan/β-1,6-glucan-containing polysaccharides (MGCP)

Commensal fungi constitute about 2% of human microbial biomass[85] and play a key role in immune regulation.[86] High-throughput sequencing techniques have revealed that the gut microbiome harbors over 50 genera of fungi, such as *Candida*, *Saccharomyces*, and *Cladosporium* species being among the most prevalent.[87] Fungal dysbiosis is increasingly recognized as a key feature of IBD.[88–90] Enhanced colonization of the intestine by *Candida* species and elevated production of anti-*Saccharomyces cerevisiae* antibodies, have been observed in patients with IBD.[91–93] Interestingly, the immunomodulatory properties of beta-glucans appear to vary based on their chemical structure, exhibiting either pro-inflammatory or anti-inflammatory effects. Under steady-state conditions, polysaccharides containing β-1,3-glucan predominantly enhance pro-inflammatory responses.[94] In contrast, a relatively less abundant class of cell surface polysaccharides, obtained from the fractionation of yeast cell wall components coupled with the enzymatic removal of β-1,3-glucan termed MGCP, has been shown to exert strong anti-inflammatory effects on the immune system.[27] These MGCPs exhibit immunomodulatory properties by promoting the induction of Tregs while simultaneously suppressing the differentiation of IFN-γ-producing Th1 cells[27] (Figure 2). Mechanistically, MGCP mediates Treg induction through the modulation of DCs in a Dectin-1-dependent manner and induces them to produce Cox2. Although Dectin-1 is traditionally associated with pro-inflammatory immune responses,[95] our data suggest that it may function in a ligand-specific manner when interacting with MGCP, thereby promoting the generation of immunoregulatory Tregs. Intriguingly, the suppressive effect of MGCP on Th1 differentiation was found to be dependent on TLR2 signaling in DCs, as TLR2-deficient DCs failed to inhibit Th1 differentiation when co-cultured with MGCP and naïve CD4$^+$ T cells. In vivo, MGCP demonstrated therapeutic potential by suppressing the progression of T-cell transfer colitis and experimental

autoimmune encephalomyelitis (EAE), underscoring its ability to mitigate inflammatory and autoimmune conditions.

However, given that Treg potentiation can hinder anti-tumor immunity, we observed that MGCP treatment exacerbated tumor growth in a mouse melanoma model. These findings provide critical insights into the complex interplay between fungal-derived polysaccharides and the host immune system, with implications for both autoimmune diseases and cancer immunotherapy. Furthermore, they underscore the importance of characterizing multiple ligands derived from microbial structural components. Identification of MGCP reveals that seemingly opposing immunomodulatory ligands may coexist within the same microbe,[27] potentially exerting their effects in a context-dependent manner to fine-tune immune responses.

Bacterial metabolites in treg generation and function

Short-chain fatty acids (SCFAs)

SCFAs are small organic molecules composed of fewer than six carbon atoms, primarily produced through the microbial fermentation of dietary fibers in the colon.[96,97] The most prominent SCFAs – acetate (C2), propionate (C3), and butyrate (C4) – play essential roles in Treg development, expansion, and function.[98,99] A comprehensive list of gut microbiota species associated with SCFA production is provided in Table 1.

SCFAs serve as key signaling molecules between gut microbiota and host immune cells. They act as ligands for G-Protein coupled receptors (GPCRs) – GPR41, GPR43, GPR109A, and Olfr78,[119–121] and induce pTreg generation and proliferation. Studies on human GPRs reveal that propionate activates both GPR43 and GPR41, acetate predominantly targets GPR43, and butyrate exhibits selectivity for GPR41.[122] Additionally, GPR109A, is specifically activated by butyrate and the vitamin niacin.[123] GPR43 is coupled to both Gαi and Gαq proteins, activating phospholipase C, inhibiting adenylyl cyclase, and triggering intracellular calcium release.[122] In Tregs, GPR43 signaling enhances mTOR activity and glycolysis, supporting cellular proliferation and functional fitness.[124] GPR41, predominantly coupled to Gαi, inhibits cAMP production and activates ERK1/2 and p38 MAPK pathways.[125] SCFA binding to GPR109A activates Gαi proteins to inhibit adenylyl cyclase and reduce cAMP levels.[126] In dendritic cells, GPR109A signaling induces the expression of anti-inflammatory genes and promotes the production of retinoic acid and IL-10, creating a tolerogenic environment conducive to Treg differentiation.[127]

In the GF mice, oral supplementation with SCFAs, significantly increased the frequency of colonic Tregs.[99] This effect was mediated through SCFA binding to GPR43, followed by inhibition of histone deacetylase (HDAC) activity,[99] leading to enhanced acetylation at *Foxp3* gene locus. In an adoptive transfer model of T cell mediated colitis, *GPR43*$^{-/-}$ CD4^{+} T cells failed to convert to Tregs upon treatment with SCFAs.[99] However, conflicting evidence exists regarding the dependency on GPCRs for SCFA-mediated Treg modulation. For instance, Park et al. demonstrated that SCFA-induced Treg differentiation occurs independently of GPR41 or GPR43 but instead relies on direct HDAC inhibition.[128] Notably, acetate, despite being a potent GPR43 agonist, failed to enhance pTreg differentiation. Further, butyrate can bind to GPR109A on colonic APCs and induce expression of *Il10* and *Aldh1a1* to induce differentiation of Tregs.

In the cell intrinsic manner, SCFAs can act as epigenetic regulator and were shown to inhibit HDAC activity and thus, enhance histone acetylation of *Foxp3* gene locus. This epigenetic modification enhances the accessibility of transcriptional machinery to promoter regions and conserved non-coding sequences (CNSs), such as CNS3, within the *Foxp3* locus. Chloroform-resistant microbial strains, including *Clostridium* species, were found to restore colonic Treg numbers in GF mice, an effect attributed to their robust production of butyrate. Indeed, dietary supplementation with butyrylated starch ameliorated CD4^{+} T cell-induced transfer colitis by enhancing colonic Treg generation. Butyrate enhanced histone H3 acetylation at both promoter and CNS3 of the *Foxp3* gene locus.[31] Similar observations were reported by Arpaia et al., showing that oral butyrate potentiates colonic pTreg differentiation via

Table 1. Bacterial strains involved with production of short-chain fatty acids.

S.No.	SCFA	Phylum	Species	Strain	References
1	Acetate	Verrucomicrobiota	*Akkermansia muciniphila*	ATCC BAA-835	Zhuge et al.[100]; Lakshmanan et al.[101]
2		Actinobacteriota	*Bifidobacterium longum*	JCM 1217	Fukuda et al.[102]; Yoon et al.[103]
3		Actinobacteriota	*Bifidobacteria adolescentis*	L2–32	O'Riordan et al.[104]; Rios-Covian et al.[105]
4		Firmicutes	*Blautia hydrogenotrophica*		Martin et al.[106]
5		Bacteroidetes	*Bacteriodes spp*		O'Riordan et al.[104]
6	Propionate	Bacteroidetes	*Bacteroides xylanisolvens*	GGCC_0124	
7		Verrucomicrobiota	*Akkermansia muciniphila*	ATCC BAA-835	
8		Verrucomicrobiota	*Akkermansia sp.*	GGCC_0220	
9		Bacteroidetes	*Bacteroides uniformis*	GGCC_0301	
10		Bacteroidetes	*Barnesiella sp.*	GGCC_0306	van der Lelie et al.[107]
11		Bacteroidetes	*Bacteroides massiliensis*	DSM 17,679	
12		Bacteroidetes	*Bacteroides stercoris*	DSM 19,555	
13		Bacteroidetes	*Barnesiella intestinihominis*	DSM 21,032	
14		Firmicutes	*Megamonas hypermegale*	DSM 1672	
15		Bacteroidetes	*Bacteroides thetaiotamicron*	VPI-5482/ATCC 29,148	O'Riordan et al.[104]; Wang et al.[108]
16		Firmicutes	*Faecalibacterium prausnitzii*	ATCC 27,766	Rios-Covian et al.[105]; Zhou et al.[109]
17		Bacteroidetes	*Bacteroides fragilis*		
18		Firmicutes	*Clostridium ramosum*		O'Riordan et al.[104]
19		Bacteroidetes	*Prevotella copri*		
20		Firmicutes	*Eubacterium rectale*	ATCC 33,656	Mukherjee et al.[110]
21		Firmicutes	*Megamonas funiformis*	DSM 19,343	
22	Butyrate	Bacteroidetes	*Bitterella massiliensis*	GGCC_0305	
23		Firmicutes	*Clostridium symbiosum*	GGCC_0272 ATCC 14,940	
24		Firmicutes	*Eubacterium callanderi*		
25		Firmicutes	*Intestinimonas butyriciproducens*	GGCC_0179	
26		Firmicutes	*Clostridium butyricum*	GGCC_0151	
27		Firmicutes	*Blautia producta*	DSM 2950	
28		Firmicutes	*Anaerostipes hadrus*	ATCC 29,173	
29		Firmicutes	*Anaerostipes caccae*	DSM 14,662	
30		Firmicutes	*Subdoligranulum variabile*	DSM 15,176	
31		Firmicutes	*Faecalibacterium prausnitzii*	DSM 17,677	van der Lelie et al.[107]; O'Riordan et al.[104]
32		Firmicutes	*Acidaminococcus intestini*	DSM 21,505	van der Lelie et al.[107]
33		Firmicutes	*Clostridium tyrobutyricum*		
34		Firmicutes	*Roseburia intestinalis*		O'Riordan et al.[104]
35		Firmicutes	*Roseburia inulinovorans*		
36		Firmicutes	*Eubacterium hallii*		
37		Firmicutes	*Eubacterium rectale*	ATCC 33,656	O'Riordan et al.[104]; Lu et al.[111]

SCFAs. Butyrate and propionate, but not acetate, increased histone acetylation on intronic CNS1 of *Foxp3* gene via HDAC inhibition.[30]

Beyond their HDAC-inhibitory effects, SCFAs can promote Treg differentiation through metabolic reprogramming. For example, propionate treatment in patients with multiple sclerosis (MS) enhanced mitochondrial oxygen consumption rates, altered mitochondrial morphology, and boosted the suppressive functionality of Tregs. This treatment also increased the proportion of circulating Tregs, contributing to the mitigation of disease progression.[129] These findings highlight the multifaceted mechanisms by which SCFAs modulate Treg biology, acting through both epigenetic and metabolic pathways (Figure 3).

In human studies, abnormal concentrations of SCFAs have been observed in various disease states, providing important clinical correlates to mechanistic findings in animal models. Patients with IBD consistently show reduced fecal SCFA levels, particularly butyrate, compared to healthy controls[130–132] correlating with impaired Treg induction, increased mucosal inflammation, and disease exacerbation. This reduction correlates with decreased abundance of butyrate-producing bacteria such as *Faecalibacterium prausnitzii* and *Roseburia* species.[133] Similarly,

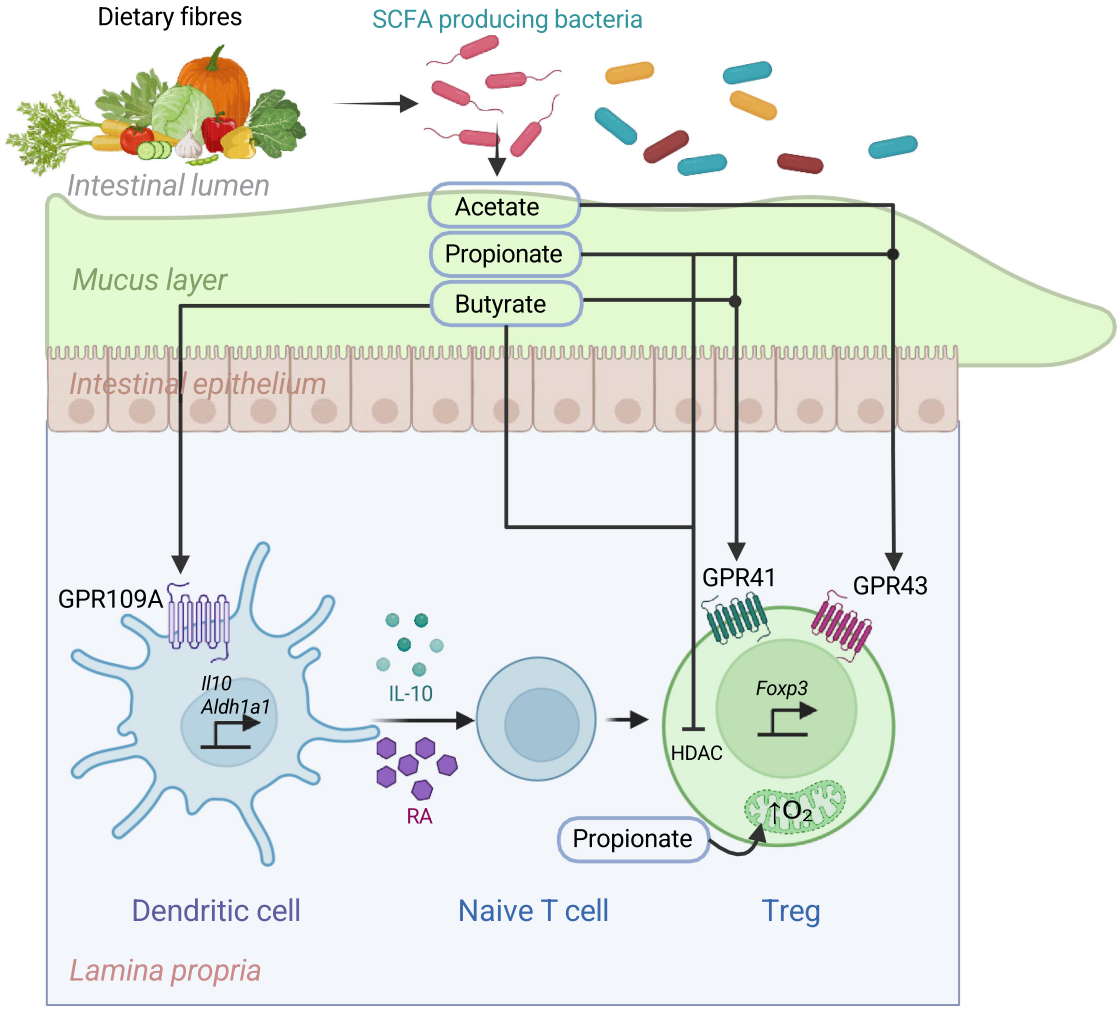

Figure 3. Mechanisms of intestinal treg modulation by microbial short-chain fatty acids (SCFAs). Gut bacteria metabolize dietary fibers to produce short-chain fatty acids (SCFAs), including acetate, propionate, and butyrate, which play a critical role in the generation and function of intestinal peripheral regulatory T cells (pTregs). Acetate: Acts as a ligand for the GPR43 receptor and inhibits histone deacetylase (HDAC) activity in Tregs upon binding to GPR43. This inhibition stabilizes Foxp3 expression, thereby enhancing the suppressive function of pTregs. Propionate: Binds to both GPR41 and GPR43 receptors and similarly inhibits HDAC activity, contributing to the stabilization of Foxp3 expression. Propionate can also diffuse into Tregs to directly inhibit HDACs. Additionally, propionate enhances oxygen consumption in Treg mitochondria, improving their metabolic fitness and functional stability. Butyrate: Functions as a ligand for GPR109A, expressed on gut dendritic cells (DCs). Activation of GPR109A upregulates the expression of *Il10* and *Aldh1a1* in DCs, leading to increased production of IL-10 and retinoic acid (RA). These factors promote the differentiation of naïve $CD4^+$ T cells (Tn) into pTregs. Butyrate can also diffuse into Tregs to inhibit HDACs, further stabilizing Foxp3 expression.

reduced SCFA levels have been reported in patients with multiple sclerosis,[134] type 1 diabetes,[135] and asthma,[136] suggesting a common metabolic signature across multiple immune-mediated conditions. In mouse models, SCFA depletion in DSS-induced colitis exacerbates inflammation and reduces Treg populations, while butyrate supplementation ameliorates symptoms.[131] Similarly, in murine CRC models, low butyrate promotes tumor growth, and in metabolic disease models, SCFA reductions impair insulin sensitivity.[6] Interestingly, there are notable discrepancies between mouse models and human conditions regarding SCFA metabolism and effects. Owing to their fiber-rich diets and *Clostridia*-enriched microbiota, mice generate higher SCFA concentrations, while typical human diets produce substantially lower SCFA levels.[6,32,131,132] Additionally, the distribution and expression patterns of SCFA receptors differ between mice and humans, potentially affecting downstream signaling pathways.[137] For instance, GPR41 and GPR43 expression patterns in immune cells show species-specific differences, which may influence the immunomodulatory effects of SCFAs.[138] These discrepancies highlight the importance of validating findings from mouse models in human studies and considering species-specific differences when translating basic research into clinical applications.

The impact of SCFAs on immune regulation appears to be highly context-dependent, with potentially divergent outcomes based on the local immune environment, concentration, and disease context.[139,140] While SCFAs are widely recognized for their immunoregulatory functions by promoting Treg differentiation and function, their immunomodulatory effects can vary based on concentration, receptor engagement, and the local immune environment.[140] Acetate has been shown to have limited effects on Treg differentiation compared to butyrate and propionate, and in some contexts, it may enhance pro-inflammatory responses by promoting effector T cell functions.[141–143] High concentrations of butyrate can induce apoptosis in colonic epithelial cells, potentially compromising barrier integrity.[144,145] Also, while promoting Treg differentiation in healthy contexts, butyrate can enhance oxidative stress and exacerbate inflammation in CRC by activating oncogenic Wnt/β-catenin signaling.[146] Similarly, propionate amplifies Treg suppressive capacity in autoimmunity but may impair anti-tumor immunity by dampening CD8+ T cell responses.[147]

Furthermore, in certain neurological conditions, elevated SCFA levels have been associated with microglial activation and neuroinflammation, highlighting their dual nature.[148] In EAE, studies have reported both protective and exacerbating effects of SCFA supplementation, suggesting complex regulatory mechanisms that may vary by disease stage and immunological context.[134,149] These findings highlight the dose- and context-dependent duality of microbial metabolites, necessitating careful therapeutic targeting.

Tryptophan metabolites

Tryptophan (Trp) is an essential aromatic amino acid for humans supplied by dietary proteins. The gut microbiome possesses diverse enzymes capable of processing dietary nutrients into a broad spectrum of metabolites, which could play an important role in host pathophysiology.[150] Despite Trp being the least abundant amino acid in proteins and cells, it is a precursor to a wide variety of microbial and host metabolites.[151] Dietary Trp is absorbed primarily in the small intestine and is metabolized through three major pathways. Approximately 90% of Trp is metabolized via Kynurenine pathway by host IDOs and tryptophan 2,3-dioxygenase (TDO) enzymes.[152,153] This generates several biologically active metabolites like kynurenine (Kyn), kynurenic acid (Kna), 3-hydroxykynurenine (3-OHKyn), 3-hydroxyanthranilic acid (3HAA), and quinolinic acid.[154] About 5% of Trp is used to synthesize serotonin by tryptophan hydroxylases (TPH1 and TPH2). Serotonin is further metabolized into melatonin through sequential enzymatic steps involving serotonin-N-acetyltransferase and acetylserotonin O-methyltransferase.[113] Notably, 90–95% of serotonin resides in the gastrointestinal tract,

predominantly within enterochromaffin cells.[112,155,156] The remaining 5% of dietary tryptophan is catabolized by gut bacteria into indole and its derivatives, including indole-3-acetic acid (IAA), indole-3-propionic acid (IPA), and others via Indole pathway.[157] This process is particularly prominent in the distal colon, as gradual depletion of carbohydrates from proximal to distal colon shifts bacterial metabolism toward protein fermentation.[158] Additionally, certain bacterial species, such as *Lactobacilli*, can degrade Trp in the stomach and ileum of mice.[159]

Serotonin

Serotonin (5-HT) has emerged as a critical mediator of immune regulation, particularly in the context of Tregs. Unlike T effector cells (Teffs), Tregs express key components of the serotonergic system, including the serotonin transporter (SERT), serotonin receptors (5-HT1a and 5-HT2), and enzyme tryptophan hydroxylase, which converts Trp into serotonin.[160] While under microbial influence, the majority of serotonin is produced by enterochromaffin cells in the gut epithelium,[155] certain bacterial species like *Streptococcus* spp., *Enterococcus* spp., *Escherichia* spp., *Lactobacillus plantarum*, *Klebsiella pneumonia*, and *Morganella morganii* – can also synthesize serotonin.[161–163] A comprehensive list of bacterial species involved in serotonin biosynthesis is provided in Table 2.

In adult GF mice, serum and plasma levels of serotonin are significantly reduced, with the most pronounced deficits observed in the colon rather than the small intestine,[115,186] suggesting a specific role of microbiota in regulating colonic 5-HT.[112] However, recent studies have revealed that during early life, gut bacteria play a dominant role in serotonin production in the small intestine. For instance, *Rodentibacter heylii* and *Enterococcus gallinarum* contribute to serotonin synthesis in mice, while *Staphylococcus aureus*, *Clostridium perfringens*, *Klebsiella grimontii*, *Staphylococcus epidermis*, and *Enterobacter cloacae* perform similar functions in human small intestine.[187] Mechanistically, this 5-HT inhibits mTORC1 in T cells via indole-3-acetaldehyde (I3A), promoting their differentiation into Tregs rather than effector T cells[187] (Figure 4). Thus, bacterial serotonin facilitates the establishment of immune tolerance to dietary antigens and commensal microbes during early perinatal development. Oral administration of serotonin to neonatal mice followed by ovalbumin (OVA) sensitization induced long-term tolerance to OVA. Moreover, T cells from serotonin-treated mice exhibited enhanced tolerogenic properties in an adoptive transfer colitis model. Interestingly, serotonin treatment also altered gut microbiota composition, suggesting bidirectional regulation between the microbiome and Tregs via serotonin signaling.[187]

However, role of serotonin and Treg interaction in immune pathology remains complex and context-dependent. In arthritic mice deficient in serotonin, there is a shift toward Th17 cell polarization.[188] Similarly, mice lacking enzyme Tph exhibit reduced Treg frequencies and increased Th17 responses during collagen-induced arthritis, effects that can be reversed by serotonin supplementation.[189] However, in humans with allergic rhinitis, elevated serum serotonin levels correlate negatively with peripheral Treg frequencies, highlighting potential discrepancies between murine models and human disease states.[190]

Indoles

Intestinal bacteria can convert the tryptophan into indole by enzyme tryptophanase (TnaA).[191] Interestingly, in mammals, indole is produced exclusively through bacterial metabolism, as host cells lack the metabolic ability to synthesize it.[192] While TnaA expression was earlier thought to be solely a characteristic of prokaryotes, recent evidence suggests that lateral gene transfer has enabled certain eukaryotic organisms, such as the gut-associated parasite *Blastocystis*, to acquire bacterial-derived TnaA,[193] which could help its adaptation to gut environment.[194]

Beyond indole, the intestinal microbiota generates a diverse array of indole-related metabolites through tryptophan catabolism. These include indole-3-pyruvate, indole-3-lactate, indole-3-propionate, indole-3-acetate, indole-3-acetamide, indole-3-acrylate, indole acetaldehyde, indole-3-aldehyde, 3-methyl-indole (skatole), and indole-3-acetaldehyde.[195,196] These metabolites play critical

Table 2. Gut bacterial strains involved in modulating gut serotonin.

S.No.	Phylum	Species	Strain	Role in Serotonin Production/Modulation	References
1	Firmicutes	*Clostridium* spp.		Stimulates enterochromaffin cells to produce serotonin via metabolites.	Yano et al.[112]
2	Firmicutes	*Lactobacillus plantarum*	Strain WCFS1	Modulates serotonin levels via tryptophan metabolism.	O'Mahony et al.[113]
3	Firmicutes	*Lactobacillus reuteri*	Strain ATCC PTA 6475	Influences serotonin levels through immune modulation.	O'Mahony et al.[113]
4	Actinobacteria	*Bifidobacterium infantis*	Strain 35,624	Modulates serotonin levels via immune modulation and tryptophan metabolism.	Desbonnet et al.[114]
5	Proteobacteria	*Escherichia coli*	Strain K-12	Produces serotonin directly by metabolizing tryptophan.	Wikoff et al.[115]
6	Firmicutes	*Enterococcus* spp.		Influences serotonin levels via metabolites affecting enterochromaffin cells.	Reigstad et al.[116]
7	Bacteroidetes	*Bacteroides* spp.		Produces short-chain fatty acids (SCFAs) that stimulate serotonin production in enterochromaffin cells.	Fukumoto et al.[117]
8	Firmicutes	*Streptococcus* spp.		May modulate serotonin levels through unclear mechanisms.	Lyte[118]

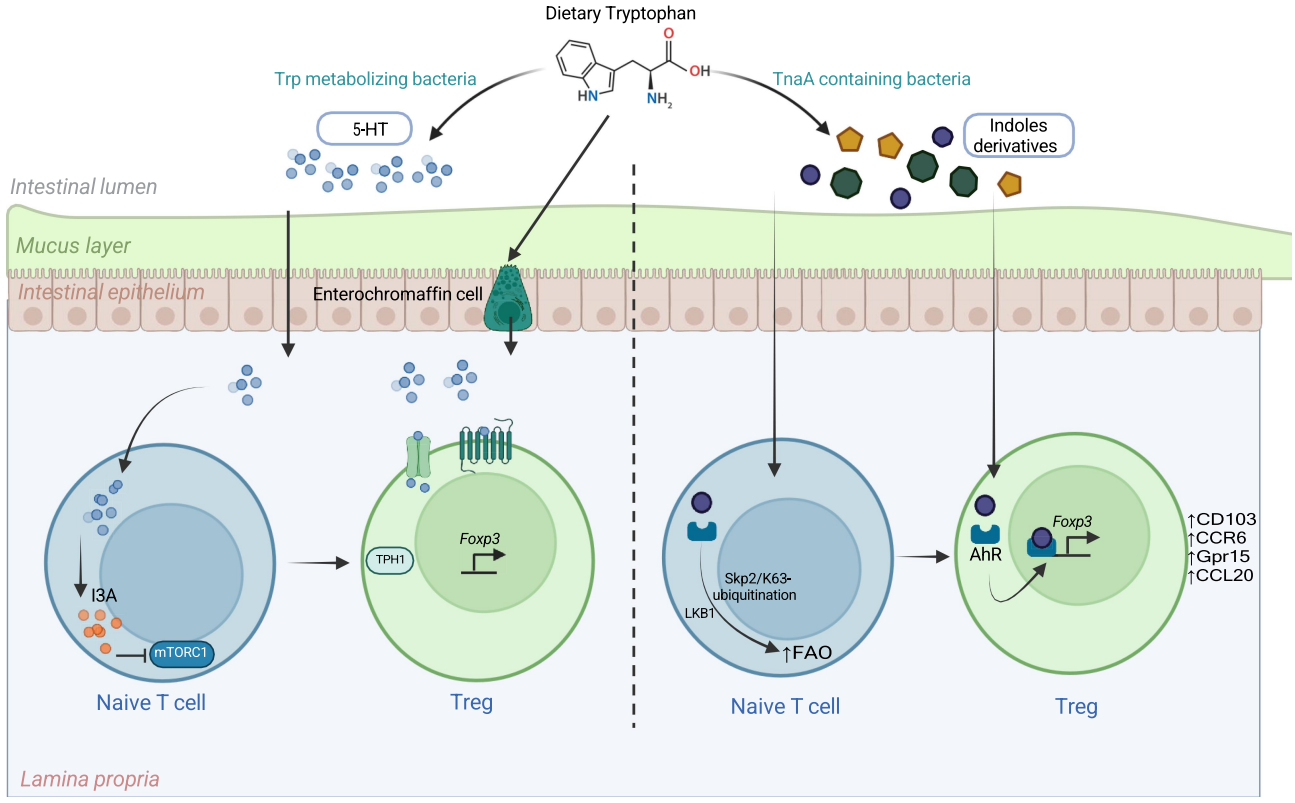

Figure 4. Mechanisms of intestinal treg modulation by microbial tryptophan metabolites. Dietary tryptophan is metabolized by gut bacteria through distinct pathways, generating bioactive compounds that modulate intestinal regulatory T cells (Tregs). Serotonin Pathway: Bacteria containing tryptophan hydroxylase convert dietary tryptophan into serotonin (5-HT). Naïve $CD4^+$ T cells take up serotonin and metabolize it into indole-3-acetaldehyde (I3A). I3A inhibits the mechanistic target of rapamycin complex 1 (mTORC1), promoting the differentiation of naïve T cells (Tn) into peripheral regulatory T cells (pTregs). Notably, Tregs themselves express serotonin receptors, transporters, and tryptophan-metabolizing enzymes, such as tryptophan hydroxylases (TPH1 and TPH2), suggesting a direct role for serotonin in Treg biology. Indole Derivatives Pathway: Bacteria expressing tryptophanase (TnaA) catabolize tryptophan into various indole derivatives in the colon. These indole metabolites serve as ligands for the aryl hydrocarbon receptor (AhR). Activation of AhR enhance Liver kinase B1 mediated fatty acid oxidation via Skp2/K63-ubiquitination pathway in $CD4^+$ T cells promoting Treg generation (left panel). Further, AhR activation enhances Foxp3 expression and gut homing molecules like CD103, CCR6, Gpr15, and CCL20 in peripheral Tregs. It reinforces the suppressive regulatory functions of Tregs, further promoting immune tolerance in the gut.

roles in maintaining intestinal barrier integrity, protecting against pathogens, and modulating host metabolism, primarily through the activation of the transcription factor aryl hydrocarbon receptor (AhR).[159,197,198] A comprehensive list of bacterial species generating indole derivatives by Trp catabolism is provided in Table 3.

AhR is expressed across multiple T cell subsets, with particularly high levels observed in Th17 cells, $FOXP3^+$ Tregs, and Tr1 cells. Intriguingly, gut-resident Tregs exhibit elevated AhR expression compared to Tregs in other tissues, underscoring its specialized role in maintaining intestinal homeostasis and regulating gut Treg functions.[211] Ahr activation affects Treg and Th17 development in a ligand-specific manner. For instance, 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD), a xenobiotic AhR ligand, promotes Treg differentiation, while 6-formylindolo[3,2-b]carbazole (FICZ), an endogenous ligand derived from indole-3-acetaldehyde (I3AA) via bacterial metabolism, drives Th17 polarization.[212–214] Thus, indole-mediated Treg differentiation and accumulation can be context-dependent and ligand-specific. Multiple AhR ligands can promote Treg development, leading to increased Treg numbers and improved outcomes in experimental autoimmune diseases.[215] AhR activation enhances the expression of gut-homing molecules such as CD103, CCR6, Gpr15, and CCL20 in peripheral Tregs, facilitating their recruitment to the intestinal mucosa (Figure 4). Although AhR-deficient Tregs retain Foxp3

Table 3. Gut bacterial strains producing indole derivatives by tryptophan catabolism.

S.No.	Bacteria Phylum	Species	Indole Metabolite	References
1	Firmicutes	*Clostridium perfringens*	Indole	
2	Firmicutes	*Clostridium bifermentans*		
3	Firmicutes	*Clostridium tertium*		
4	Firmicutes	*Clostridium septicum*		
5	Firmicutes	*Clostridium histolyticum*		
6	Firmicutes	*Clostridium ramosum*		
7	Firmicutes	*Clostridium innocuum*		
8	Firmicutes	*Clostridium baratii*		
9	Firmicutes	*Clostridium paraputrificum*		
10	Firmicutes	*Clostridium beijerinckii*		
11	Firmicutes	*Clostridium acetobutylicum*		
12	Firmicutes	*Enterococcus faecalis*		
13	Firmicutes	*Clostridium sporogenes*		
14	Firmicutes	*Clostridium difficile*		
15	Firmicutes	*Clostridium butyricum*		
16	Bacteroidetes	*Bacteroides thetaiotaomicron*		Smith and Macfarlane[164]; Lee and Lee[165]; Buffie et al.[166]; Chen et al.[167]; Elsden et al.[168]; Devlin et al.[169]
17	Bacteroidetes	*Bacteroides ovatus*		
18	Firmicutes	*Clostridium limosum*		
19	Firmicutes	*Clostridium bifermentans*		
20	Firmicutes	*Clostridium malenomenatum*		
21	Firmicutes	*Clostridium lentoputrescens*		
22	Firmicutes	*Clostridium tetani*		
23	Firmicutes	*Clostridium tetanomorphum*		
24	Firmicutes	*Clostridium ghoni*		
25	Firmicutes	*Clostridium sordellii*		
26	Proteobacteria	*Desulfovibrio vulgaris*		
27	Firmicutes	*Enterococcus faecalis*		
28	Proteobacteria	*Escherichia coli*		
29	Fusobacteriota	*Fusobacterium nucleatum*		
30	Proteobacteria	*Haemophilus influenza*		
31	Firmicutes	*Peptostreptococcus asscharolyticus*		
32	Firmicutes	*Faecalibacterium prausnitzii*	Indole-3-propionic acid (IPA)	Sokol et al.[170]

(*Continued*)

Table 3. (Continued).

S.No.	Bacteria Phylum	Species	Indole Metabolite	References
33	Firmicutes	*Lactobacillus reuteri*	Indole-3-lactic acid (ILA)	Zelante et al.[159]; Smith and Macfarlane[164]; Aragozzini et al.[171]; Cervantes-Barragan et al.[172]; Dodd et al.[173]; Honore et al.[174]; Russell et al.[175]; Wilck et al.[176]
34	Firmicutes	*Lactobacillus plantarum*		
35	Firmicutes	*Lactobacillus casei*		
36	Firmicutes	*Lactobacillus acidophilus*		
37	Firmicutes	*Anaerostipes hadrus*		
38	Firmicutes	*Anaerostipes caccae*		
39	Bacteroidetes	*Bacteroides thetaiotaomicron*		
40	Bacteroidetes	*Bacteroides eggerthii*		
41	Bacteroidetes	*Bacteroides ovatus*		
42	Bacteroidetes	*Bacteroides fragilis*		
43	Actinobacteriota	*Bifidobacterium adolescentis*		
44	Actinobacteriota	*Bifidobacterium bifidum*		
45	Actinobacteriota	*Bifidobacterium longum* subsp. *infantis*		
46	Actinobacteriota	*Bifidobacterium longum* subsp. *longum*		
47	Actinobacteriota	*Bifidobacterium pseudolongum*		
48	Firmicutes	*Clostridium bartlettii*		
49	Firmicutes	*Clostridium perfringens*		
50	Firmicutes	*Clostridium sporogenes*		
51	Firmicutes	*Clostridium saccharolyticum*		
52	Proteobacteria	*Escherichia coli*		
53	Firmicutes	*Eubacterium rectale*		
54	Firmicutes	*Eubacterium cylindroides*		
55	Firmicutes	*Faecalibacterium prausnitzii*		
56	Firmicutes	*Lactobacillus murinus*		
57	Firmicutes	*Lactobacillus paracasei*		
58	Firmicutes	*Lactobacillus reuteri*		
59	Firmicutes	*Megamonas hypermegale*		
60	Bacteroidetes	*Parabacteroides distasonis*		
61	Firmicutes	*Peptostreptococcus asscharolyticus*		

(*Continued*)

Table 3. (Continued).

S.No.	Bacteria Phylum	Species	Indole Metabolite	References
62	Firmicutes	*Ruminococcus gnavus*	Indole-3-acetic acid (IAA)	
63	Firmicutes	*Roseburia spp.*		
64	Firmicutes	*Coprococcus comes*		
65	Firmicutes	*Blautia spp.*		
66	Firmicutes	*Clostridium scindens*		
67	Firmicutes	*Clostridium bartlettii*		
68	Firmicutes	*Clostridium hiranonis*		
69	Firmicutes	*Clostridium hylemonae*		
70	Firmicutes	*Clostridium sordellii*		
71	Bacteroidetes	*Bacteroides thetaiotaomicron*		
72	Bacteroidetes	*Bacteroides eggerthii*		
73	Bacteroidetes	*Bacteroides ovatus*		
74	Bacteroidetes	*Bacteroides fragilis*		
75	Actinobacteriota	*Bifidobacterium adolescentis*		
76	Actinobacteriota	*Bifidobacterium longum* subsp. *longum*		
77	Actinobacteriota	*Bifidobacterium pseudolongum*		Smith et al.,[164] Elsden et al.,[168] Russell et al.[175]; Li et al.[177]; Barbeyron et al.[178]; Valles-Colomer et al.,[179]; Zhu et al.[180]; Spanogiannopoulos et al.[181]
78	Firmicutes	*Clostridium bartlettii*		
79	Firmicutes	*Clostridium difficile*		
80	Firmicutes	*Clostridium lituseburense*		
81	Firmicutes	*Clostridium paraputrificum*		
82	Firmicutes	*Clostridium perfringens*		
83	Firmicutes	*Clostridium putrefaciens*		
84	Firmicutes	*Clostridium saccharolyticum*		
85	Firmicutes	*Clostridium sticklandii*		
86	Firmicutes	*Clostridium subterminale*		
87	Proteobacteria	*Escherichia coli*		
88	Firmicutes	*Eubacterium hallii*		
89	Firmicutes	*Eubacterium cylindroides*		
90	Bacteroidetes	*Parabacteroides distasonis*		
91	Firmicutes	*Peptostreptococcus asscharolyticus*		

(*Continued*)

Table 3. (Continued).

S.No.	Bacteria Phylum	Species	Indole Metabolite	References
92	Bacteroidetes	*Bacteroides thetaiotaomicron*	3-methylindole (Skatole)	
93	Firmicutes	*Butyrivibrio fibrisolvens*		
94	Firmicutes	*Clostridium bartlettii*		
95	Firmicutes	*Clostridium scatologenes*		
96	Firmicutes	*Clostridium drakei*		Russell et al.[175]; Honeyfield et al.[182]; Whitehead et al.[183]
97	Firmicutes	*Eubacterium cylindroides*		
98	Firmicutes	*Eubacterium rectale*		
99	Firmicutes	*Lactobacillus* spp.		
100	Firmicutes	*Megamonas hypermegale*		
101	Bacteroidetes	*Parabacteroides distasonis*		
102	Firmicutes	*Clostridium sporogenes*	Indoleacrylic acid (IA)	
103	Firmicutes	*Peptostreptococcus russellii*		Dodd et al.[173]; Wlodarska et al.[184]
104	Firmicutes	*Peptostreptococcus anaerobius*		
105	Firmicutes	*Peptostreptococcus stomatis*		
106	Firmicutes	*Lactobacillus acidophilus*	Indolealdehyde (IAld)	
107	Firmicutes	*Lactobacillus murinus*		Zelante et al.[159]; Cervantes-Barragan et al.[172]; Wilck et al.[176]
108	Firmicutes	*Lactobacillus reuteri*		
109	Firmicutes	*Clostridium botulinum*	Indolepropionic acid (IPA)	
110	Firmicutes	*Clostridium caloritolerans*		
111	Firmicutes	*Clostridium paraputrificum*		
112	Firmicutes	*Clostridium sporogenes*		
113	Firmicutes	*Clostridium cadvareris*		Wikoff et al.[115]; Elsden et al.[168]; Dodd et al.[173]; Wlodarska et al.[184]; Williams et al.[185]
114	Firmicutes	*Peptostreptococcus asscharolyticus*		
115	Firmicutes	*Peptostreptococcus russellii*		
116	Firmicutes	*Peptostreptococcus anaerobius*		
117	Firmicutes	*Peptostreptococcus stomatis*		
118	Firmicutes	*Clostridium sporogenes*	Tryptamine	Williams et al.[185]
119	Firmicutes	*Ruminococcus gnavus*		

expression, they lose their suppressive functionality, emphasizing the critical role of AhR in Treg-mediated immune regulation.[216] Interestingly, Ahr expression in intestinal Tregs is not dependent on microbiota, as GF or antibiotic-treated mice show no differences in Treg AhR levels.[216]

A phytochemical AhR ligand, indigo naturalis, has been shown to promote the accumulation of Helios$^+$ Tregs near MHCII$^+$ epithelial cells in intestinal crypts, further supporting the role of AhR in shaping the gut immune landscape.[217] Additionally, AhR ligands enhance Liver kinase B1 mediated fatty acid oxidation via Skp2/K63-ubiquitination pathway in CD4$^+$ T cells promoting Treg generation (Figure 4), which protect mice from DSS-induced colitis.[218] In its inactive state, AhR resides in the cytoplasm as part of a complex with heat shock protein 90 (HSP90), AhR-interacting protein (AIP), and p23.[219] Upon binding to ligands AhR undergoes conformational changes that expose its nuclear localization signal, leading to translocation into the nucleus.[213] In the nucleus, AhR dimerizes with the AhR nuclear translocator (ARNT) and binds to specific DNA sequences known as xenobiotic response elements (XREs) in the promoter regions of target genes.[220] In Tregs, AhR activation induces the expression of genes involved in Treg differentiation and function, including Foxp3, IL-10, and TGF-β.[221] Additionally, AhR can interact with other transcription factors, such as c-Maf, to synergistically enhance IL-10 production.[222] Furthermore, AhR activation in dendritic cells induces the expression of IDO1, creating a feedback loop that enhances kynurenine production and further activates AhR signaling.[223]

The interplay between microbial indole derivatives and Tregs remains an emerging area of research. A recent study demonstrated that the probiotic *Lactobacillus reuteri*, a producer of indole-3-lactate, cross-feeds other bacterial species and enhances microbial tryptophan metabolism.[224] Elevated production of indole derivatives enriches the gut microbiota with *Clostridium* clusters XIVa, XIVb, and IV, known inducers of colonic Tregs.[225] This microbial shift confers protection against *Citrobacter rodentium* infection and alleviates DSS-induced colitis.[224]

Conversely, disruptions in microbial indole metabolism can impair immune tolerance. Stephen-Victor et al. recently revealed that goblet-cell-derived resistin-like molecule β (RELMβ) influences the gut microbiome by depleting indole-metabolite-producing bacteria like *Lactobacilli* and *Alistipes*.[226] This is achieved through the upregulation of antimicrobial genes such as *Sprr2a1/2/3* and *Reg3*, which alters the microbial balance, impairs oral tolerance, and exacerbates food allergy responses. *Lactobacilli* produce indole derivatives like IAA, I3A, and ILA, which promote the expansion of RORγt$^+$ Tregs via AhR activation. In a mouse model of IL-4 receptor gain-of-function-induced food allergy, reintroducing *Lactobacilli* restored oral tolerance, whereas deleting AhR in Tregs abolished this protective effect.[226] In conclusion, microbial indole derivatives and their interaction with AhR represent a critical axis in regulating intestinal immunity and Treg function. These metabolites not only shape the composition of the gut microbiota but also influence immune homeostasis and disease susceptibility. While significant progress has been made in elucidating the roles of indoles and AhR in immune regulation, further research is needed to fully unravel the intricate mechanisms underlying these interactions. Such insights hold immense therapeutic potential for modulating gut immunity and treating inflammatory and autoimmune disorders.

In humans, abnormal tryptophan metabolism has been observed in various inflammatory and autoimmune conditions. Patients with IBD show reduced serum levels of tryptophan and altered kynurenine pathway metabolites like indole-3-aldehyde, indicating enhanced IDO1 activity.[227] Similarly, patients with multiple sclerosis exhibit altered tryptophan metabolism, with changes in the kynurenine-to-tryptophan ratio correlating with disease activity.[228] Recent metabolomic studies have also identified reduced levels of AhR ligands in patients with psoriasis[229] and rheumatoid arthritis,[230] suggesting impaired tryptophan metabolism by the gut microbiota. Significant differences exist between mice and humans regarding tryptophan metabolism and AhR signaling. The affinity of various tryptophan metabolites for the AhR differs between species, with some ligands showing high potency in mice but limited activity in humans.[231] Additionally, the

expression patterns of enzymes involved in tryptophan metabolism vary between species, affecting the spectrum of metabolites produced.[159] These differences may explain some of the challenges in translating AhR-targeted therapies from mouse models to human diseases. Future studies should focus on identifying human-specific AhR ligands and understanding their role in immune regulation to develop more effective therapeutic strategies.

Bile acids

Bile acids (BAs) are amphipathic metabolites derived from cholesterol in the liver and play a crucial role in the digestion and absorption of dietary fats. Beyond their classical functions in lipid metabolism, BAs are now recognized as critical regulators of glucose and energy homeostasis.[232] Further, identification of their receptors has paved the way for a deeper understanding of their hormone-like characteristics in regulating immune homeostasis.[233]

In humans, the liver synthesizes two primary BAs: cholic acid (CA) and chenodeoxycholic acid (CDCA). In contrast, rodents produce additional muricholic acids (MCA), which are 6-hydroxylated derivatives of CDCA.[234] These primary BAs are conjugated with glycine or taurine in the liver before being secreted into the duodenum.[235,236] Approximately 95% of secreted BAs are reabsorbed in terminal ileum and recycled back to liver via enterohepatic circulation. The remaining BAs enter the colon, where they undergo extensive microbial transformation.

Gut microbiota possessing bile salt hydrolase activity, such as bacteria from the genera *Lactobacillus*, *Bifidobacterium*, *Clostridium*, and *Bacteroides*, are able to deconjugate the BAs by cleaving the glycine or taurine moiety attached to the steroid core.[232,234] Deconjugated BAs are further modified through dehydroxylation, epimerization, oxidation, desulfation, esterification, and reconjugation. For example, the dehydroxylation of CA and CDCA at the C7 position generates secondary BAs, including DCA and lithocholic acid (LCA), respectively. In mice, murideoxycholic acid is also formed from MCA.[237] GF animals lack secondary BAs, underscoring the essential role of gut microbiota in bile acid metabolism.[238,239] A list of gut bacterial strains involved in BA transformation reactions is provided in Table 4. BA-metabolizing enzymes help bacteria to overcome BA toxicity. Conversely, BAs help sustain microbial diversity, with human tauro-β-MCA and taurocholic acid playing key roles in shaping an adult-like microbiome.[240] Dysregulation of bile acid metabolism, as seen in cholestasis or bile acid ligation models, is associated with reduced microbial diversity.[241,242]

BAs exert their immunomodulatory effects via a heterogenous family of transmembrane GPCRs and nuclear receptors. The nuclear receptor farnesoid X receptor (FXR) serves as the primary receptor for CDCA in humans and CA in mice,[243–245] while secondary BAs like DCA and LCA activate G-protein bile acid receptor 1 (GBPAR1, also known as Takeda G-protein receptor (TGR5).[246] Additionally, DCA and LCA interact with other nuclear receptors, including the vitamin D receptor (VDR),[247] pregnane-X-receptor (PXR),[248] and constitutive androstane receptor (CAR).[249] Emerging evidence also implicates muscarinic M3 receptors[250] and sphingosine-1-phosphate receptor[251] in BA signaling (Figure 5).

Several studies have highlighted critical role of BAs and their derivatives in regulating Treg differentiation in the intestine. Two derivatives of LCA – 3-oxoLCA and isoalloLCA – generated by bacterial modification of primary BAs have been shown to modulate T cell differentiation. 3-oxoLCA inhibits the differentiation of Th17 cells by directly binding to transcription factor RORγt. IsoalloLCA, on the other hand, increased mitochondrial reactive oxygen species (mtROS) leading to enhanced FOXP3 expression utilizing CNS3 enhancer region in the *Foxp3* locus.[32] IsoalloLCA also promotes histone acetylation at the *Foxp3* promoter in the presence of TGF-β signaling (Figure 5).[32] Subsequent work identified *Bacteroidetes* species as producers of isoalloLCA and demonstrated that its induction of mtROS generates Tregs via activation of nuclear receptor NR4A1 (Figure 5).[252] Notably, patients with IBD exhibit reduced representation of genes encoding enzymes for isoalloLCA production in gut microbiome, along

Table 4. Gut bacterial strains involved in bile acid transformation reactions.

S.No.	Phylum	Species	Strain	Reference
1	Actinobacteria	*Bifidobacterium adolescentis*		Lucas et al.[199]
2	Actinobacteria	*Bifidobacterium bifidum*		
3	Actinobacteria	*Bifidobacterium dentium*		
4	Actinobacteria	*Collinsella aerofaciens*		
5	Actinobacteria	*Collinsella intestinalis*		
6	Actinobacteria	*Collinsella stercoris*		
7	Bacteroidetes	*Alistipes indistinctus*		
8	Bacteroidetes	*Bacteroides caccae*		
9	Bacteroidetes	*Bacteroides finegoldii*		
10	Bacteroidetes	*Bacteroides intestinalis*		
11	Bacteroidetes	*Bacteroides ovatus*		
12	Bacteroidetes	*Bacteroides thetaiotaomicron*	3731	
13	Bacteroidetes	*Bacteroides thetaiotaomicron*	7330	
14	Bacteroidetes	*Bacteroides thetaiotaomicron*	VPI-5482	
15	Bacteroidetes	*Bacteroides uniformis*		
16	Bacteroidetes	*Bacteroides vulgatus*		
17	Bacteroidetes	*Bacteroides xylanisolvens*		
18	Firmicutes	*Blautia hansenii*		
19	Firmicutes	*Blautia luti*		
20	Firmicutes	*Clostridium asparagiforme*		
21	Firmicutes	*Clostridium hylemonae*		
22	Firmicutes	*Clostridium leptum*		
23	Firmicutes	*Clostridium* M62_1		
24	Firmicutes	*Clostridium scindens*		
25	Firmicutes	*Coprococcus comes*		
26	Firmicutes	*Dorea formicigenerans*		
27	Firmicutes	*Dorea longicatena*		
28	Firmicutes	*Enterocloster bolteae* (formerly *Clostridium*)		
29	Firmicutes	*Erysipelatoclostridium ramosum* (formerly *Clostridium*)		
30	Firmicutes	*Holdemania filiformis*		
31	Firmicutes	*Hungatella hathewayi* (formerly *Clostridium*)		
32	Firmicutes	*Lactobacillus ruminis*		
33	Firmicutes	*Roseburia intestinalis*		
34	Firmicutes	*Ruminococcus* GM2/1		
35	Firmicutes	*Ruminococcus gnavus*		
36	Firmicutes	*Ruminococcus torques*		
37	Firmicutes	*Tyzzerella nexilis* (formerly *Clostridium nexile*)		
38	Fusobacterium	*Fusobacterium varium*		
39	Proteobacteria	*Escherichia coli*	K12 MG1655	
40	Proteobacteria	*Escherichia fergusonii*		
41	Proteobacteria	*Proteus penneri*		
42	Firmicutes	*Lactobacillus plantarum*	K21	Wu et al.[200]
43	Firmicutes	*Clostridium scindens*	ATCC 35,704	Ridlon et al.[201]; Wahlstrom et al.[202]
44	Actinomycetota	*Eggerthella lenta*		Doden et al.[203]
45	Firmicutes	*Ruminococcus gnavus*	ATCC 29,149	Doden et al.[203]
46	Firmicutes	*Bacillus subtilis*	R0179	Culpepper et al.[204]
47	Actinomycetota	*Bifidobacterium animalis subsp. lactis*	B94	Culpepper et al.[204]
48	Bacteroidetes	*Bacteroides fragilis*	NCTC 9343, ATCC 25,285	Sun et al.[205]
49	Firmicutes	*Lactobacillus salivarius*		Xu et al.[206]
50	Firmicutes	*Lactobacillus plantarum*	WCFS1, ATCC14197	Prete et al.[207]
51	Firmicutes	*Lactobacillus acidophilus*	ATCC 4356	Wu et al.[208]
52	Actinomycetota	*Eggerthella lenta*	DSM 2243, C592	Harris et al.[209]
53	Bacteroidetes	*Bacteroides thetaiotaomicron*	VPI-5482, ATCC 25,285	Adhikari et al.[210]
54	Bacillota	*Eubacterium rectale*	ATCC 33,656	Mukherjee et al.[110]

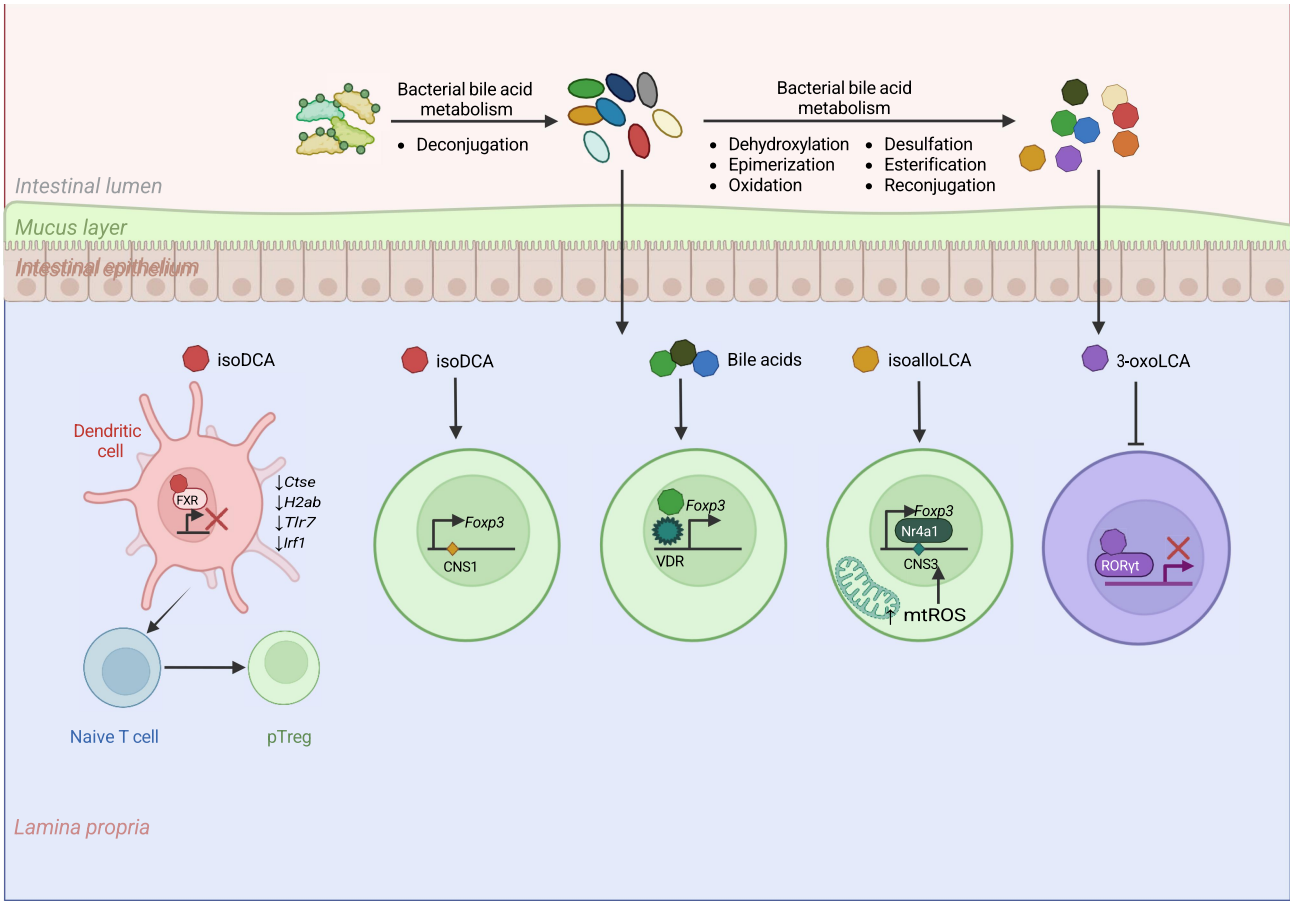

Figure 5. Mechanisms of intestinal treg modulation by microbial bile acid Metabolites. Gut bacteria play a key role in bile acid (BA) metabolism by deconjugating primary bile acids that escape the enterohepatic circulation. These deconjugated BAs undergo further modifications by gut microbiota, generating secondary BAs that modulate intestinal Treg responses through diverse mechanisms. isoDCA : This secondary bile acid metabolite inhibits the nuclear farnesoid X receptor (FXR) in dendritic cells (DCs), promoting their tolerogenic phenotype. These tolerogenic DCs facilitate the differentiation of naïve $CD4^+$ T cells into $CD4^+RORγt^+Foxp3^+$ pTregs (pTregs). Additionally, isoDCA enhances *Foxp3* expression in pTregs through a CNS1-mediated mechanism, further stabilizing their regulatory function. Bile Acids and Vitamin D Receptor (VDR) : Certain bile acid metabolites act via the nuclear vitamin D receptor to upregulate *Foxp3* expression, driving the generation of pTregs and reinforcing immune tolerance. isoalloLCA: A metabolite derived from lithocholic acid (LCA), isoalloLCA increases mitochondrial reactive oxygen species (ROS) in Tregs. This leads to the activation of the transcription factor Nr4a1 which enhances *Foxp3* expression through a CNS3-dependent mechanism, strengthening Treg suppressive activity. 3-oxoLCA: Another LCA-derived metabolite, 3-oxoLCA, suppresses Th17 cell differentiation by inhibiting RORγt binding, thereby reducing pro-inflammatory Th17 responses.

with decreased microbial synthesis of this metabolite.[252] Human gut bacteria *Gordonibacter pamelaeae* P7-E3, *Eggerthella lenta* P7-G7, *Raoultibacter massiliensis* P7-A2, *Collinsella intestinalis* P8-C1, *Adlercreutzia equolifaciens* P11-C8 and *Clostridium citroniae* P2-B6 were later identified as top converters of LCA to 3-oxoLCA.[253]

BAs also expand pTregs through interactions with their receptors. Campbell et al. discovered that the secondary BA 3β-hydroxydeoxycholic acid (isoDCA) induces an anti-inflammatory phenotype in DCs by inhibiting the FXR activity, thereby promoting pTreg differentiation.[254] The interaction between isoDCA and FXR downregulated several pro-inflammatory genes involved in antigen processing, presentation, and pro-inflammatory signal transduction in DCs (Figure 5). Furthermore, bacteria engineered to produce isoDCA enhanced colonic $RORγt^+$ pTregs in a CNS1-dependent manner.[254] Primary and secondary BAs can also induce $RORγt^+$ pTregs by interacting with Treg-intrinsic VDR (Figure 5).[255] This effect does not rely on Vitamin D3, as colonic $RORγt^+$ pTregs were unaffected by its absence in diet but were significantly reduced by Treg-specific VDR deletion.

Overall, BAs, gut microbiota, and colonic pTregs form a dynamic and interdependent network essential for establishing intestinal immune tolerance. Intestinal BAs are indispensable for maintaining colonic pTregs, while gut microbes are instrumental in shaping this relationship by metabolizing BAs. Dysregulation of this triadic interaction can disrupt immune tolerance, contributing to inflammatory diseases such as IBD. Indeed, administration of BAs like LCA[255] or rationally designed consortium composed of BA-producing bacteria[107] have shown promise in reducing colitis severity.

Human studies have revealed significant alterations in bile acid profiles across various disease states. Patients with IBD show increased levels of primary bile acids and decreased secondary bile acids in feces, reflecting impaired microbial bile acid metabolism.[256] This dysregulation is particularly pronounced in Crohn's disease patients with ileal involvement, where bile acid malabsorption contributes to diarrhea and other symptoms.[257,258] Similarly, patients with primary sclerosing cholangitis, which is often associated with IBD, exhibit distinct bile acid signatures characterized by elevated levels of toxic bile acids.[259] Notable species differences exist in bile acid metabolism between mice and humans. Mice produce muricholic acids, which are potent FXR antagonists, whereas these bile acids are absent in humans.[260] Additionally, the gut microbiota composition differs substantially between mice and humans, affecting the spectrum of secondary bile acids produced.[261] These differences may explain some of the discrepancies observed when translating findings from mouse models to human conditions. For instance, while certain bile acid receptor agonists show promising results in mouse models of colitis, their efficacy in human IBD has been variable.[262] Understanding these species-specific differences is crucial for developing targeted therapies based on bile acids for human diseases.

Additionally, microbial metabolites such as secondary BAs can have context-dependent effects, with some derivatives promoting inflammation under specific conditions.[263] For instance, DCA has been implicated in pro-inflammatory responses in certain disease states, potentially exacerbating liver inflammation and colorectal cancer progression by inducing DNA damage.[264,265] Furthermore, indole derivatives, while activating AhR-dependent Treg pathways, can also drive Th17 polarization in the presence of pro-inflammatory cytokines like IL-6.[197] These findings highlight the dose- and context-dependent duality of microbial metabolites, necessitating careful therapeutic targeting.

Impact of impaired immune responses on microbiota

While the influence of the microbiota on immune function has been extensively studied, the reciprocal impact of impaired immune responses on microbiota composition and function is equally important but less well characterized. Defects in Treg function or number can significantly reshape the intestinal microbial landscape, creating a dysbiotic environment that may further exacerbate immune dysregulation.[5] Studies in mice with specific immune deficiencies have provided valuable insights into this relationship. For instance, mice lacking the anti-inflammatory cytokine IL-10, which is crucial for Treg function, develop spontaneous colitis accompanied by significant alterations in their gut microbiota, including increased abundance of pro-inflammatory Proteobacteria and decreased levels of beneficial Firmicutes.[266] Similarly, Foxp3-deficient mice, which lack functional Tregs, exhibit profound dysbiosis characterized by the expansion of mucosa-associated segmented filamentous bacteria and other potentially pathogenic species.[267,268]

In humans, primary immunodeficiencies affecting Treg development or function, such as IPEX (Immune dysregulation, polyendocrinopathy, enteropathy, X-linked) syndrome caused by FOXP3 mutations, are associated with significant alterations in gut microbiota composition.[269] These patients often exhibit reduced microbial diversity and increased abundance of opportunistic pathogens, which may contribute to their gastrointestinal symptoms and systemic inflammation. Beyond genetic immunodeficiencies, acquired impairments in immune function can also impact the microbiota. For example, HIV infection, which depletes $CD4^+$ T cells including Tregs, leads to significant dysbiosis characterized by increased pathobiont abundance and reduced levels of beneficial

bacteria.[270] Similarly, immunosuppressive therapies used in transplantation and autoimmune diseases can alter the gut microbiota composition, potentially contributing to opportunistic infections and other complications.[271]

The mechanisms by which impaired immune responses affect the microbiota are multifaceted. Defects in antimicrobial peptide production, mucus layer integrity, and IgA secretion – all of which can be influenced by Treg function – directly impact microbial colonization and composition.[272] Additionally, alterations in cytokine profiles and intestinal inflammation can create selective pressures that favor the expansion of certain bacterial species over others.[273] This bidirectional relationship creates a potential feedback loop: impaired immune function leads to dysbiosis, which further exacerbates immune dysregulation, potentially contributing to chronic inflammation and disease pathogenesis. Understanding this complex interplay is crucial for developing targeted interventions that restore both immune homeostasis and a healthy microbiota.

Dysregulation of microbiome-treg axis in diseases

Inflammatory bowel disease (IBD)

Microbial dysbiosis and metabolite alterations in IBD

Dysbiosis, characterized by alterations in the diversity, composition, and function of the gut microbiota is a key aspect of IBD. The relationship between dysbiosis and IBD remains complex and bidirectional making it challenging to ascertain whether dysbiosis is a cause or consequence of the disease. Nonetheless, studies on GF mouse models have demonstrated that IBD either fails to develop or is significantly attenuated in the absence of gut microbes, underscoring the critical role of the microbiome in the pathogenesis of IBD.[274] Genome-wide association studies have found that many of genomic loci associated with IBD are responsible for host-microbiome interactions.[275,276] Gut bacterial -diversity significantly decreases in both ulcerative colitis (UC) and Crohn's disease (CD) forms of IBD.[277,278] Multi-omics[33] and multi-biome[13] analysis have revealed a consistent depletion of obligate anaerobes like *Faecalibacterium prausnitzii* and *Roseburia hominis*[13,33,279] among other SCFA-producing bacteria like *Eubacterium* spp. (*E. rectale* and *E. ventriosum*), *Blautia* spp., *Bacteroides* spp., and *Anaerostipes hadrus*. Indeed, the IBD metabolome presented with a general reduction in SCFAs.[33] Additionally, this was accompanied by a significant reduction in *Subdoligranulum* sp., which forms a complex of new species-level clade with at least seven butyrate producer species of *Subdoligranulum*, *Gemmiger*, and *Faecalibacterium* genera.[170,280]

An increase in primary bile acid cholate and its glycine and taurine conjugates was also observed in CD patients while secondary BAs lithocholate and deoxycholate were reduced.[33] This shift suggests a depletion of secondary BA-producing bacteria or faster colonic transit times that limit microbial BA transformation in IBD patients.[256,281] Additionally, an increase in fungal diversity has been reported in both UC and CD.[282] IBD patients display an increased abundance of *Candida albicans* and a decreased abundance of *Saccharomyces cerevisiae*. However, *S. cerevisiae* was found enriched in a CD cohort in Japan and USA but was depleted in the China cohort.[13] This suggests a geographical heterogeneity effect on IBD-associated mycobiome. Nevertheless, high levels of anti-*Saccharomyces cerevisiae* antibodies are robust biomarkers of CD.[278,283]

Treg dysfunction and therapeutic implications in IBD

Although IBD has a complex pathophysiology with the involvement of multiple factors, these findings indicate that dysbiosis-induced Treg dysfunction may play a role in IBD in genetically susceptible individuals, as both SCFAs and BAs are important for maintaining Treg-mediated gut immune tolerance. Indeed, colonization of GF mice with human fecal microbiota from IBD patients resulted in an increased number of Th17 cells and a reduced population of RORγt^{+} Tregs, compared to mice colonized with microbiota from healthy donors.[284] Paradoxically, in human patients of CD colon lamina propria, Tregs are enriched while the circulating Tregs are decreased during active disease.[285–287] Although Tregs present in the intestinal mucosa of IBD patients continue to express activation markers such as CTLA-4 and

PD-1,[285,288] these cells exhibit functional impairments and fail to effectively suppress inflammation.[286,289] Notably, while Tregs derived from the mucosa of CD patients retain the ability to suppress peripheral $CD4^+$ Teff cells isolated from blood, they are unable to exert similar suppressive effects on mucosal Teffs. Further, this finding suggests that gut-resident Teffs acquire resistance to Treg-mediated suppression during active IBD.[290] Comprehensive single-cell analyses of intestinal tissues from various human IBD cohorts have uncovered distinct Treg subsets within the inflamed mucosa. These subsets exhibit a spectrum of Foxp3 expression and produce proinflammatory cytokines such as IL-17 and IFN-γ. Notably, a memory-like $IL\text{-}17^+$ Treg population has been identified in patients with UC,[291] alongside a TNF^+ Treg subset,[292] which might contribute to the anti-TNF treatment resistance in IBD patients.

Thus, schemes to expand functional mucosal Tregs or enhance their function can provide protection from IBD. Indeed, Treg expansion therapies like low-dose IL-2 treatment have been shown to provide moderate clinical response in UC patients with significant expansion of Tregs.[293] Similarly, recent studies have demonstrated that microbial restoration through fecal microbiota transplantation (FMT) can improve outcomes in patients with UC form of IBD.[294–296] Additionally, a defined consortium of probiotics, selected for their ability to produce beneficial metabolites such as SCFAs, indoles, and bile salts[107] has demonstrated efficacy in ameliorating experimental colitis in murine models. This probiotic consortium not only reversed dysbiosis but also restored a functional gut microbiome capable of generating anti-inflammatory metabolites associated with mucosal homeostasis. Furthermore, it enhanced protective immunity by significantly increasing the frequency of IL-10-producing $ROR\gamma t^+$ $FoxP3^+$ Tregs. While microbe-derived products like PSA, CSGG, and MGCP have shown promising results in resolving experimental colitis in mice, clinical data remain limited. Nevertheless, given their ability to induce Tregs, it is reasonable to hypothesize that administrating these bioactive compounds from beneficial bacteria (postbiotics) could elicit favorable therapeutic responses in human IBD, warranting further investigation in clinical trials.

Celiac disease (CeD)

Immune dysregulation and treg dysfunction in CeD

CeD is a chronic hyperimmune disorder caused by an abnormal immune response to gliadin, a component of gluten, in genetically predisposed individuals. Having compatible human leukocyte antigen (HLA) genetics is necessary for the development of CeD, but it alone does not cause the condition. While around 40% of the population possesses the permissive HLA genes, only approximately 3% of individuals develop CeD during their lifetime.[297] This highlights the critical role of additional genetic, environmental, and immunological factors in disease pathogenesis. Though associated with changes in gut bacteria, a consistent microbial signature in patients has not been identified.[298] The pathogenesis of CeD is known to primarily mediated by gluten-specific inflammatory Th1 and Th17 cells.[299,300] Multiple studies have reported simultaneous expression of regulatory cytokines like IL-10 and TGF-β along with inflammatory cytokines IFN-γ, IL-17, and IL-21 in CeD.[301–303] This creates a paradoxical environment in untreated CeD, where regulatory mechanisms attempt to suppress inflammation and mitigate the abnormal immune response triggered by gliadin.[304]

Studies have revealed intriguing parallels between CeD and IBD regarding Treg dynamics, as CeD is also characterized by an increase in $Foxp3^+$ Tregs in small intestinal lamina propria.[305,306] However, their suppressive functions are impaired significantly.[304,307,308] IL-15 is significantly overexpressed in the intestines of celiac patients, where it contributes to immune dysfunction by disrupting TGF-β signaling, impairing Treg activity, and rendering Teff cells resistant to Treg-mediated suppression through activation of PI3K pathway.[309,310] Additionally, Serena et al.[311] highlighted the role of gut microbiome in the hypofunction of Tregs in CeD. In active CeD, the loss of intestinal barrier integrity allows microbial-derived

butyrate to synergize with IFN-γ to modulate alternative splicing of *FOXP3*, favoring the expression of shorter *FOXP3* Delta 2 isoform, which lacks exon 2. This isoform compromises the interaction between FOXP3 and transcription factors RORαt and RORγt, thereby promoting Th17 differentiation.[312] This shift in *FOXP3* isoform expression underscores how the intestinal microenvironment can reprogram Tregs, undermining their capacity to maintain immune tolerance and exacerbating the inflammatory response in CeD.

Microbiota alterations in CeD

The CeD-associated microbiota changes have been studied in high-risk infants with a first-degree relative diagnosed with CeD. These studies have revealed distinct microbial signatures, with increased abundance of the *Bacteroides-Prevotella* group,[313] *Firmicutes*, *Proteobacteria*, and *Bifidobacterium* in infants compared to controls.[314] Another study found that such infants exhibit a lower abundance of *Bacteroides* and a higher abundance of *Firmicutes* compared to healthy controls.[315] In a longitudinal study, Olivares et al.[16] observed that children who later developed CeD showed an increased abundance of *Firmicutes*, particularly *Enterococcaceae* and *Peptostreptococcaceae*, between 4 and 6 months of age. In contrast, no such differences were observed in control individuals during the same period. These findings suggest that early-life microbial dysbiosis may precede and potentially contribute to CeD pathogenesis.

A recent ongoing prospective clinical trial,[14] utilizing shotgun metagenomic sequencing for functional characterization of microbes, Celiac Disease Genomic, Environmental, Microbiome and Metabolome study (CDGEMM), has further elucidated the relationship between environmental factors and microbial changes in high-risk infants. The study found that formula feeding was associated with an increased abundance of *Ruminococcus gnavus* and *Lachnospiraceae bacterium*, both of which have been linked to allergic and inflammatory conditions. Additionally, infants delivered by cesarean section exhibited a decreased abundance of *Bacteroides vulgatus* and *Bacteroides dorei*, alongside broader metabolomic alterations. One particularly intriguing finding from the CDGEMM study was the decreasing abundance of propionic acid in high-risk infants. Propionic acid is a known inducer of functionally competent Tregs.[129] While it remains to be determined whether these microbial and metabolic changes directly contribute to CeD development, these findings underscore the potential importance of restoring Treg functionality or modulating the gut microbiome as novel therapeutic strategies.

Future research should focus on unraveling the precise mechanisms by which microbial and environmental factors influence immune regulation in CeD. Understanding these pathways could pave the way for innovative interventions aimed at restoring durable immune tolerance and preventing disease onset in genetically predisposed individuals.

Colorectal cancer

Microbial alterations in CRC

Colorectal cancers (CRCs) are intrinsically linked to the gut microbiota due to their anatomical location within the gastrointestinal tract. Transplanting fecal microbiota from CRC patients into GF mice promotes colonic cell proliferation and accelerates colon tumor formation. Conversely, fecal microbiota from cancer-free individuals do not have the same effect, underscoring the role of CRC-associated microbiota in disease progression.[316] A comprehensive multi-cohort metagenomic analysis identified a core bacterial signature of seven CRC-enriched bacterial species—*Bacteroides fragilis*, *Fusobacterium nucleatum*, *Porphyromonas asaccharolytica*, *Parvimonas micra*, *Prevotella intermedia*, *Alistipes finegoldii*, and *Thermanaerovibrio acidaminovorans*—that were consistently present across diverse populations spanning various geographies and ethnicities.[317] In addition to these CRC-associated bacteria, the study also identified 62 bacterial species that were depleted in CRC patients. Notably, five of these depleted species—*Clostridium butyricum*, *Streptococcus salivarius*, *Streptococcus thermophilus*, *Carnobacterium maltaromaticum*, and *Lactobacillus gallinarum*—have been associated with health-promoting effects, underscoring their potential protective roles in the context of CRC development. Further, gut bacteria have been shown to modify response to

immune check-point inhibitor therapy in multiple tumor types,[318–320] including CRC.[321–323] Fecal metagenomic and metabolomic data from individuals at various stages of colorectal tumorigenesis revealed dynamic changes in gut microbes and metabolites from early adenoma to the late stage of CRC suggesting dysbiotic changes could be drivers of CRC tumorigenesis.[324]

F. nucleatum ssp. nucleatum, *Solobacterium moorei*, *Peptostreptococcus stomatis*, *Peptostreptococcus anaerobius*, *Lactobacillus sanfranciscensis*, *Parvimonas micra*, and *Gemella morbillorum* are bacterial species that increased across all stages of tumor progression, while *Atopobium parvulum*, *Actinomyces odontolyticus*, *Desulfovibrio longreachensis*, and *Phascolarctobacterium succinatutens* were elevated only in early stages. Two butyrate-producing bacteria *Lachnospira multipara*, and *Eubacterium eligens* are significantly depleted in CRC. While relatively less studied, this loss of beneficial bacteria can be instrumental in CRC tumorigenesis. Furthermore, in the early stages of CRC, there is an increase in bile salt DCA, glycocholate, and taurocholate, indicating a role in tumorigenesis.[324] Indeed, DCA increases DNA damage and mutations,[325] while administration of BAs increases gut tumor incidences in mice.[326]

Tregs in CRC progression and therapy

Chronic inflammation is a well-established risk factor for the development and progression of various cancers, including CRC.[327] The role of Tregs in this context presents a complex relationship with tumor progression. While they are pivotal in maintaining immune homeostasis and suppressing exuberant inflammation under normal conditions, their increased presence in tumors is implicated in cancer progression and indicates a worsening prognosis.[25,35] Studies demonstrate that Tregs adopt a hyper-suppressive phenotype within TME, actively suppressing anti-tumor immunity and thus promoting CRC progression.[328,329] These findings align with our recent demonstration that CRC-infiltrating Tregs exhibit enhanced activation of the NF-κB subunit C-REL, a Treg-effector transcription factor, driven by increased post-translational *O*-GlcNAcylation, which may contribute to their heightened immunosuppressive functions.[330] However, some studies have reported that elevated densities of $Foxp3^+$ Tregs correlate with suppression of CRC progression.[331,332] These apparent contradictions may be explained by the heterogeneity of cells expressing FOXP3 in humans. Saito et al.[333] identified a subset of $FOXP3^{lo}CD45RA\text{-}CD4^+$ TILs that transiently express FOXP3 but lack the canonical suppressive functions of bona fide Tregs. These cells are characterized by high expression of proinflammatory cytokines such as IL-17 and IFN-γ, suggesting that their accumulation in CRC may enhance anti-tumor immunity rather than suppress it and thus, their accumulation in CRC accentuates the anti-tumor immunity.[333]

Furthermore, considering the signature microbiota, which is depleted in the initiation stages of CRC being instrumental in colonic differentiation of $ROR\gamma t^+$ Tregs and activation of colonic Tregs of thymic origin, it is highly probable that Tregs maintain a low inflammatory environment in the gut promoting intestinal immune homeostasis and thus, potentially inhibiting the tumorigenesis in the gut. Supporting this notion, a recent study by Frei et al.[334] spatially resolved the immune markers over 3,000 CRC samples, distinguishing between intraepithelial and intrastromal compartments. Strikingly, they found that higher densities of intraepithelial $CD8^+$ T cells and intrastromal $Foxp3^+$ Tregs were strongly predictive of favorable clinical outcomes. The association of better prognosis with intrastromal rather than intraepithelial Tregs underscores their potential role in controlling inflammation and limiting tumor invasiveness. These findings suggest that enhancing the frequency and functionality of colonic Tregs through targeted interventions, such as specific probiotics, postbiotics, live biotherapeutic products, or microbial-derived ligands, could represent a promising therapeutic strategy for CRC. Characterizing the unique markers and mechanisms of stromal Tregs that inhibit tumor growth will be crucial for developing precise microbiome-based therapies. Such approaches could harness the immunoregulatory properties of Tregs to maintain gut immune homeostasis while simultaneously mitigating chronic inflammation, thereby offering a dual benefit in CRC prevention and treatment. Further research into the interplay between the gut microbiota, Treg biology, and tumor microenvironment dynamics will pave the way for innovative strategies aimed at modulating Treg activity to improve patient outcomes in CRC.

Despite the strong associations between microbiota alterations and various inflammatory and autoimmune diseases, establishing causality remains a significant challenge in the field. To distinguish whether dysbiosis is a cause or consequence of disease is inherently difficult due to the bidirectional nature of host-microbiome interactions.[335] Studies in GF mouse models demonstrate that the absence of microbiota attenuates disease severity in IBD, suggesting a contributory role of the microbiome.[161] Similarly, FMT from IBD patients to GF mice transfers disease phenotypes,[284] however, reverse causality where inflammation itself reshapes the microbiota complicates interpretations. For example, intestinal inflammation reduces oxygen tolerance, favoring the expansion of facultative anaerobes like Proteobacteria.[336] Moreover, clinical trials of probiotics and prebiotics have yielded mixed results - *Lactobacillus rhamnosus* GG ameliorates eczema but fails to prevent asthma,[337] while high-fiber diets improve Treg responses in some IBD cohorts but show no benefit in others.[338] Genetic polymorphisms in immune receptors (e.g., TLRs, NLRP3) further modulate individual responses to microbial signals, suggesting that microbiota-Treg interactions are heavily influenced by host factors.[276] Moreover, geographical and genetic heterogeneity in microbial signatures, as observed with Saccharomyces cerevisiae in Crohn's disease cohorts, underscores the challenge of establishing universal microbial drivers of disease.[13] Furthermore, many studies reporting microbiome alterations in disease states are cross-sectional rather than longitudinal, limiting their ability to establish temporal relationships necessary for causal inference.[339] These contradictory findings highlight the need for caution in interpreting the microbiota-Treg axis as uniformly beneficial. Future research leveraging longitudinal studies, multi-omics approaches, mechanistic studies, and controlled microbial interventions is essential to move beyond correlative observations and establish causal relationships in the microbiome-Treg axis and dissect the context-specific roles of microbial communities in immune regulation.

Clinical translation: trials and challenges in targeting the microbiota-Treg axis

The promising results from preclinical studies targeting the microbiota-Treg axis have spurred numerous clinical trials, with varying degrees of success. Understanding both the successes and failures of these trials provides valuable insights for future therapeutic development.

Low-dose IL-2 therapy has emerged as a promising approach to expand Tregs in vivo. Several clinical trials have demonstrated that low-dose IL-2 can selectively expand Tregs without significantly affecting effector T cells in patients with various autoimmune conditions.[340] In a phase 1/2 trial involving patients with ulcerative colitis, low-dose IL-2 treatment resulted in significant clinical improvement in 50% of patients, accompanied by expansion of $FOXP3^+$ Tregs.[341] However, challenges remain regarding the optimal dosing regimen, potential off-target effects, and long-term efficacy of this approach. Similarly, adoptive Treg transfer represents another strategy to restore immune tolerance. Early-phase clinical trials have demonstrated the safety and feasibility of ex vivo expanded autologous Tregs in conditions such as type 1 diabetes[342] and Crohn's disease.[343] However, a phase 1 trial of ovalbumin-specific Tregs in Crohn's disease patients failed to show significant clinical benefit despite demonstrating safety.[341]

FMT has shown promise in recurrent *Clostridioides difficile* infection and is being investigated for various immune-mediated conditions. In ulcerative colitis, several randomized controlled trials have demonstrated modest efficacy of FMT in inducing clinical remission.[344,345] A trial of FMT in Crohn's disease showed moderate benefit,[346] highlighting the disease-specific effects of this approach. The variability in donor stool composition, optimal administration protocols, and long-term safety concerns remain significant challenges for FMT.

Probiotic interventions have yielded mixed results in clinical trials. While some studies have shown modest benefits of specific probiotic strains in conditions such as ulcerative colitis,[347,348] others have failed to demonstrate significant effects as in atopic dermatitis.[349] A notable failure was the PROPATRIA trial, which found that a probiotic

mixture increased mortality in patients with severe acute pancreatitis,[350] highlighting the potential risks of untargeted microbial interventions in certain clinical contexts. Postbiotic interventions, using microbial-derived components or metabolites, represent an emerging approach with potential advantages over live bacterial therapies. Early-phase trials of SCFA supplementation[351] has shown promising effects on immune parameters, but larger efficacy trials are still needed.

However, clinical trials specifically examining the relationship between Tregs and microbial interventions remain limited, with most evidence coming from preclinical models or observational studies. Though several studies are investigating FMT in immune-mediated conditions, only few directly measured Treg outcomes. Al et al.[352] conducted a pilot randomized controlled trial of FMT in multiple sclerosis patients (NCT03183869), measuring peripheral blood cytokines as the primary outcome. While this trial demonstrated that FMT was safe and tolerable, with potential to improve intestinal permeability and enrich for an MS-protective microbiota, it did not specifically report Treg changes. Similarly, NCT02516384 examined two donor FMT in ulcerative colitis patients with immunological assessments.[353] Interestingly, along with moderate improvement in clinical response they found that both mucosal Th1 cells and Tregs were decreased post-FMT. Reduction in Tregs probably happened concomitant to reduction in mucosal inflammation as a result of increased microbial diversity. Preclinical evidence suggests that microbial interventions can influence Treg populations, as demonstrated in murine models where defined microbiota transplants restored Th17/RORγt$^+$ regulatory T cell balance, but human clinical trial data with direct Treg outcome measurements remains an important gap in the current literature.

Furthermore, oral consumption as a substitute for bacterial functionality presents both opportunities and challenges. While oral administration of bacterial metabolites like SCFAs, tryptophan derivatives, or BAs could theoretically bypass the need for a functional microbiota, several limitations exist. These include the poor stability of many metabolites in the gastrointestinal tract, challenges in achieving physiologically relevant concentrations at target sites, and the loss of context-dependent production of these metabolites.[354] Additionally, many bacterial functions involve complex metabolic networks and cell-to-cell interactions that cannot be easily replicated by single metabolites.[355] Despite these challenges, targeted delivery systems and synthetic biology approaches are being developed to overcome some of these limitations. For example, engineered bacteria designed to produce specific metabolites or immune-modulating molecules in response to environmental cues represent a promising approach to combine the advantages of live bacteria with the specificity of postbiotic interventions.[356]

The mixed results from clinical trials targeting the microbiota-Treg axis highlight the complexity of translating preclinical findings to human diseases. Future success will likely depend on more personalized approaches that consider individual variations in microbiota composition, genetic factors, and disease heterogeneity. Additionally, combination therapies that target multiple aspects of the microbiota-Treg axis may prove more effective than single interventions.

Conclusion and future perspectives

The interplay between the gut microbiome and Tregs represents a cornerstone of immune homeostasis, with profound implications for health and disease. This review has highlighted the multifaceted mechanisms by which microbial components and metabolites shape Treg development, differentiation, and function. These microbial-derived signals not only maintain intestinal immune tolerance but also influence systemic immunity, underscoring the gut microbiome's role as a key modulator of immune responses.

Dysregulation of the microbiome-Treg axis is a hallmark of inflammatory and autoimmune diseases. In IBD, microbial dysbiosis and reduced production of immunomodulatory metabolites, such as SCFAs, impair Treg function, leading to chronic inflammation.[33] Similarly, emerging evidence suggests microbiome-based changes in other conditions, such as MS and autism spectrum disorders (ASD), among others. In MS, alterations in gut microbial composition have been linked to immune dysregulation and disease progression,[357,358] while

in ASD, gut microbiome imbalances correlate with behavioral and neurological symptoms.[359] In cancer therapy, specific microbial signatures have been identified as predictors of response to immune checkpoint inhibitors, highlighting the potential for microbiome modulation to enhance treatment efficacy.[360,361] However, in many instances, it remains unclear whether microbial changes are a cause or consequence of disease processes, necessitating further investigation to establish causal relationships and mechanistic insights.

Emerging evidence suggests that targeting the microbiome-Treg axis holds immense therapeutic potential. Strategies such as FMT, probiotics, postbiotics, LBPs, and microbial-derived ligands have shown promise in preclinical and clinical studies.[107,294] However, translating these findings into effective therapies requires a deeper understanding of the complex interactions between microbial signals, host immunity, and disease-specific contexts.

From a therapeutic perspective, FMT has emerged as a well-established approach for modulating the gut microbiota and correcting dysbiosis. Indeed, FMT-related therapeutics have been approved by the US FDA for recurrent *Clostridioides difficile* infections.[362,363] However, defined consortia of bacteria offer significant advantages over FMT. These consortia can mimic the natural complexity of the gut microbiome, provide functional redundancy to ensure therapeutic stability, and promote stable colonization, potentially leading to long-term effects.[364] Moreover, they can simultaneously target multiple pathways, making them suitable for complex diseases. However, the use of live biotherapeutics presents challenges, including variable responses in heterogeneous patient populations and inconsistent efficacy outcomes, necessitating rigorous investigation and well-designed clinical trials to address these limitations.[365] Both LBP and FMT efficacy is highly context-dependent, influenced by factors such as donor and recipient microbiota composition, host immune status, host genetics, and delivery methods, which may limit long-term benefits.[366,367] For instance, FMT trials in ulcerative colitis show variable remission rates due to differences in donor microbial profiles and patient baseline microbiota which may either facilitate or inhibit colonization by the introduced strains.[368,369] Similarly, LBP outcomes, such as those with VE303, vary based on colonization success and host factors.[370] The complex ecological dynamics within the gut microbiota, including competition for nutrients and niches, cross-feeding relationships, and antagonistic interactions, further complicate the predictability of microbiota-based interventions.[371] These findings underscore the need for personalized approaches and further research to optimize donor selection, delivery protocols, and patient stratification to achieve sustained therapeutic outcomes.

In contrast, purified microbial products, such as PSA, CSGG, MGCP, RHP,[27,29,73] and other microbial-derived ligands, may offer a more controlled and precise approach. These well-defined products enable consistent outcomes and facilitate the study of precise mechanisms, providing better control over therapeutic interventions. Logistically, purified products, if they have a simple chemical structure, might be safer, easier to manufacture and store, and face fewer regulatory hurdles compared to live consortia or FMT. Despite these advantages, the exploration of microbial products is still in its infancy, and a plethora of bioactive molecules remain to be discovered for various dysbiotic diseases. Additionally, the roles of understudied components of the human microbiome other than bacteria, such as fungi and viruses, in Treg regulation warrant further investigation, as they may hold untapped therapeutic and biomarker potential.[372–374]

Future research should focus on elucidating the precise molecular mechanisms by which microbial components and metabolites modulate Treg biology. Personalized microbiome-based therapies, tailored to individual microbial and immune profiles, could improve treatment outcomes and pave the way for precision medicine in immune-mediated diseases.[375] Furthermore, the integration of multi-omics approaches, including metagenomics, metabolomics, and single-cell sequencing, will provide deeper insights into the microbiome-Treg axis and its role in health and disease.[376,377]

In conclusion, the microbiome-Treg axis represents a dynamic and bidirectional relationship that is central to immune homeostasis and disease. The context-dependent nature of microbial effects on immune regulation necessitates personalized approaches that consider individual variations in microbiota composition, host genetics, and disease pathophysiology.[378] Moreover, the complex interplay between beneficial and potentially harmful microbial signals requires careful consideration when developing microbiota-based therapeutics. As demonstrated by failed clinical trials with FMT in ulcerative colitis, not all patients respond uniformly to microbiome-targeted interventions, highlighting the need for better stratification approaches and more precise manipulation of specific microbial pathways.[345] Future research should focus on establishing causality through longitudinal studies, identifying disease-specific microbial signatures with strain-level characterization, metabolite profiling in disease-specific contexts, integration of multi-omics data, and developing targeted approaches to modulate specific aspects of the microbiome-Treg axis while minimizing unintended consequences. By unraveling the complexities of this interaction, we can harness the therapeutic potential of the microbiome to restore immune tolerance and improve outcomes in inflammatory, autoimmune, and neoplastic diseases. The development of microbiome-based therapies, whether through live consortia, purified products, or personalized interventions, holds immense promise for revolutionizing the treatment of immune-mediated disorders.

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CRediT: **Amit Sharma:** Conceptualization, Data curation, Formal analysis, Funding acquisition, Investigation, Methodology, Validation, Visualization, Writing – original draft, Writing – review & editing; **Garima Sharma:** Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Validation, Visualization, Writing – original draft, Writing – review & editing; **Sin-Hyeog Im:** Conceptualization, Funding acquisition, Resources, Supervision, Writing – original draft, Writing – review & editing.

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References

1. Arumugam M, Raes J, Pelletier E, Le Paslier D, Yamada T, Mende DR, Fernandes GR, Tap J, Bruls T, Batto J-M, et al. Enterotypes of the human gut microbiome. Nature. 2011;473(7346):174–180. doi: 10.1038/nature09944.
2. Eckburg PB, Bik EM, Bernstein CN, Purdom E, Dethlefsen L, Sargent M, Gill SR, Nelson KE, Relman DA. Diversity of the human intestinal microbial flora. Science. 2005;308(5728):1635–1638. doi: 10.1126/science.1110591.
3. Rinninella E, Raoul P, Cintoni M, Franceschi F, Miggiano GAD, Gasbarrini A, Mele MC. What is the healthy gut microbiota composition? A changing ecosystem across age, environment, diet, and diseases. Microorganisms. 2019;7(1):14. doi: 10.3390/microorganisms7010014.
4. Chakraborty N. Metabolites: a converging node of host and microbe to explain meta-organism. Front Microbiol. 2024;15:1337368. doi: 10.3389/fmicb.2024.1337368.
5. Round JL, Mazmanian SK. The gut microbiota shapes intestinal immune responses during health and disease. Nat Rev Immunol. 2009;9(5):313–323. doi: 10.1038/nri2515.
6. Lavelle A, Sokol H. Gut microbiota-derived metabolites as key actors in inflammatory bowel disease. Nat Rev

Gastroenterol Hepatol. 2020;17(4):223–237. doi: 10.1038/s41575-019-0258-z.
7. Morrison DJ, Preston T. Formation of short chain fatty acids by the gut microbiota and their impact on human metabolism. Gut Microbes. 2016;7(3):189–200. doi: 10.1080/19490976.2015.1134082.
8. Magnusdottir S, Ravcheev D, de crecy-Lagard V, Thiele I. Systematic genome assessment of B-vitamin biosynthesis suggests co-operation among gut microbes. Front Genet. 2015;6:148. doi: 10.3389/fgene.2015.00148.
9. LeBlanc JG, Milani C, de Giori GS, Sesma F, van Sinderen D, Ventura M. Bacteria as vitamin suppliers to their host: a gut microbiota perspective. Curr Opin Biotechnol. 2013;24(2):160–168. doi: 10.1016/j.copbio.2012.08.005.
10. Koppel N, Maini Rekdal V, Balskus EP. Chemical transformation of xenobiotics by the human gut microbiota. Science. 2017;356(6344). doi: 10.1126/science.aag2770.
11. Rosenberg E. Diversity of bacteria within the human gut and its contribution to the functional unity of holobionts. NPJ Biofilms Microbiomes. 2024;10(1):134. doi: 10.1038/s41522-024-00580-y.
12. Honda K, Littman DR. The microbiota in adaptive immune homeostasis and disease. Nature. 2016;535(7610):75–84. doi: 10.1038/nature18848.
13. Akiyama S, Nishijima S, Kojima Y, Kimura M, Ohsugi M, Ueki K, Mizokami M, Hattori M, Tsuchiya K, Uemura N, et al. Multi-biome analysis identifies distinct gut microbial signatures and their crosstalk in ulcerative colitis and Crohn's disease. Nat Commun. 2024;15(1):10291. doi: 10.1038/s41467-024-54797-8.
14. Leonard MM, Karathia H, Pujolassos M, Troisi J, Valitutti F, Subramanian P, Camhi S, Kenyon V, Colucci A, Serena G, et al. Multi-omics analysis reveals the influence of genetic and environmental risk factors on developing gut microbiota in infants at risk of celiac disease. Microbiome. 2020;8(1):130. doi: 10.1186/s40168-020-00906-w.
15. Ohnmacht C, Park JH, Cording S, Wing JB, Atarashi K, Obata Y, Gaboriau-Routhiau V, Marques R, Dulauroy S, Fedoseeva M, et al. The microbiota regulates type 2 immunity through RORγt + T cells. Science. 2015;349(6251):989–993. doi: 10.1126/science.aac4263.
16. Olivares M, Walker AW, Capilla A, Benitez-Paez A, Palau F, Parkhill J, Castillejo G, Sanz Y. Gut microbiota trajectory in early life may predict development of celiac disease. Microbiome. 2018;6(1):36. doi: 10.1186/s40168-018-0415-6.
17. Belkaid Y, Hand TW. Role of the microbiota in immunity and inflammation. Cell. 2014;157(1):121–141. doi: 10.1016/j.cell.2014.03.011.
18. Hepworth MR, Fung TC, Masur SH, Kelsen JR, McConnell FM, Dubrot J, Withers DR, Hugues S, Farrar MA, Reith W, et al. Group 3 innate lymphoid cells mediate intestinal selection of commensal bacteria–specific CD4 + T cells. Science. 2015;348(6238):1031–1035. doi: 10.1126/science.aaa4812.
19. Mowat AM, Agace WW. Regional specialization within the intestinal immune system. Nat Rev Immunol. 2014;14(10):667–685. doi: 10.1038/nri3738.
20. Coombes JL, Siddiqui KR, Arancibia-Carcamo CV, Hall J, Sun CM, Belkaid Y, Powrie F. A functionally specialized population of mucosal CD103+ DCs induces Foxp3+ regulatory T cells via a TGF-β– and retinoic acid–dependent mechanism. J Experiment Med. 2007;204(8):1757–1764. doi: 10.1084/jem.20070590.
21. Pabst O. New concepts in the generation and functions of IgA. Nat Rev Immunol. 2012;12(12):821–832. doi: 10.1038/nri3322.
22. Roncarolo MG, Gregori S, Bacchetta R, Battaglia M, Gagliani N. The biology of T regulatory type 1 cells and their therapeutic application in immune-mediated diseases. Immunity. 2018;49(6):1004–1019. doi: 10.1016/j.immuni.2018.12.001.
23. Rosser EC, Mauri C. Regulatory B cells: origin, phenotype, and function. Immunity. 2015;42(4):607–612. doi: 10.1016/j.immuni.2015.04.005.
24. Sakaguchi S, Yamaguchi T, Nomura T, Ono M. Regulatory T cells and immune tolerance. Cell. 2008;133(5):775–787. doi: 10.1016/j.cell.2008.05.009.
25. Sharma A, Rudra D. Emerging functions of regulatory T cells in tissue homeostasis. Front Immunol. 2018;9:883. doi: 10.3389/fimmu.2018.00883.
26. Sefik E, Geva-Zatorsky N, Oh S, Konnikova L, Zemmour D, McGuire AM, Burzyn D, Ortiz-Lopez A, Lobera M, Yang J, et al. Individual intestinal symbionts induce a distinct population of RORγ + regulatory T cells. Science. 2015;349(6251):993–997. doi: 10.1126/science.aaa9420.
27. Lee C, Verma R, Byun S, Jeun EJ, Kim GC, Lee S, Kang H-J, Kim CJ, Sharma G, Lahiri A, et al. Structural specificities of cell surface β-glucan polysaccharides determine commensal yeast mediated immuno-modulatory activities. Nat Commun. 2021;12(1):3611. doi: 10.1038/s41467-021-23929-9.
28. Mazmanian SK, Round JL, Kasper DL. A microbial symbiosis factor prevents intestinal inflammatory disease. Nature. 2008;453(7195):620–625. doi: 10.1038/nature07008.
29. Verma R, Lee C, Jeun E-J, Yi J, Kim KS, Ghosh A, Byun S, Lee C-G, Kang H-J, Kim G-C, et al. Cell surface polysaccharides of bifidobacterium bifidum induce the generation of Foxp3+ regulatory T cells. Science Immunol. 2018;3(28):eaat6975. doi: 10.1126/sciimmunol.aat6975.
30. Arpaia N, Campbell C, Fan X, Dikiy S, van der Veeken J, deRoos P, Liu H, Cross JR, Pfeffer K, Coffer PJ, et al. Metabolites produced by commensal bacteria promote peripheral regulatory T-cell generation. Nature. 2013;504(7480):451–455. doi: 10.1038/nature12726.

31. Furusawa Y, Obata Y, Fukuda S, Endo TA, Nakato G, Takahashi D, Nakanishi Y, Uetake C, Kato K, Kato T, et al. Commensal microbe-derived butyrate induces the differentiation of colonic regulatory T cells. Nature. 2013;504(7480):446–450. doi: 10.1038/nature12721.
32. Hang S, Paik D, Yao L, Kim E, Trinath J, Lu J, Ha S, Nelson BN, Kelly SP, Wu L, et al. Bile acid metabolites control TH17 and Treg cell differentiation. Nature. 2019;576(7785):143–148. doi: 10.1038/s41586-019-1785-z.
33. Lloyd-Price J, Arze C, Ananthakrishnan AN, Schirmer M, Avila-Pacheco J, Poon TW, Andrews E, Ajami NJ, Bonham KS, Brislawn CJ, et al. Multi-omics of the gut microbial ecosystem in inflammatory bowel diseases. Nature. 2019;569(7758):655–662. doi: 10.1038/s41586-019-1237-9.
34. Ivanov II II, Atarashi K, Manel N, Brodie EL, Shima T, Karaoz U, Wei D, Goldfarb KC, Santee CA, Lynch SV, et al. Induction of intestinal Th17 cells by segmented filamentous bacteria. Cell. 2009;139(3):485–498. doi: 10.1016/j.cell.2009.09.033.
35. Sakaguchi S, Mikami N, Wing JB, Tanaka A, Ichiyama K, Ohkura N. Regulatory T cells and human disease. Annu Rev Immunol. 2020;38(1):541–566. doi: 10.1146/annurev-immunol-042718-041717.
36. Korn LL, Hubbeling HG, Porrett PM, Yang Q, Barnett LG, Laufer TM. Regulatory T cells occupy an isolated niche in the intestine that is antigen independent. Cell Rep. 2014;9(5):1567–1573. doi: 10.1016/j.celrep.2014.11.006.
37. Cebula A, Seweryn M, Rempala GA, Pabla SS, McIndoe RA, Denning TL, Bry L, Kraj P, Kisielow P, Ignatowicz L. Thymus-derived regulatory T cells contribute to tolerance to commensal microbiota. Nature. 2013;497(7448):258–262. doi: 10.1038/nature12079.
38. Gaboriau-Routhiau V, Rakotobe S, Lecuyer E, Mulder I, Lan A, Bridonneau C, Rochet V, Pisi A, De Paepe M, Brandi G, et al. The key role of segmented filamentous bacteria in the coordinated maturation of gut helper T cell responses. Immunity. 2009;31(4):677–689. doi: 10.1016/j.immuni.2009.08.020.
39. Lee YK, Menezes JS, Umesaki Y, Mazmanian SK. Proinflammatory T-cell responses to gut microbiota promote experimental autoimmune encephalomyelitis. Proc Natl Acad Sci USA. 2011;108(Suppl):4615–4622. doi: 10.1073/pnas.1000082107.
40. Maeda Y, Kurakawa T, Umemoto E, Motooka D, Ito Y, Gotoh K, Hirota K, Matsushita M, Furuta Y, Narazaki M, et al. Dysbiosis contributes to arthritis development via activation of autoreactive T cells in the intestine. Arthritis Rheumatol. 2016;68(11):2646–2661. doi: 10.1002/art.39783.
41. Scher JU, Sczesnak A, Longman RS, Segata N, Ubeda C, Bielski C, Rostron T, Cerundolo V, Pamer EG, Abramson SB, et al. Expansion of intestinal prevotella copri correlates with enhanced susceptibility to arthritis. Elife. 2013;2:e01202. doi: 10.7554/eLife.01202.
42. Fan TJ, Goeser L, Lu K, Faith JJ, Hansen JJ. Enterococcus faecalis glucosamine metabolism exacerbates experimental colitis. Cellular Mol Gastroenterol Hepatol. 2021;12(4):1373–1389. doi: 10.1016/j.jcmgh.2021.06.017.
43. Lengfelder I, Sava IG, Hansen JJ, Kleigrewe K, Herzog J, Neuhaus K, Hofmann T, Sartor RB, Haller D. Complex bacterial consortia reprogram the colitogenic activity of enterococcus faecalis in a gnotobiotic mouse model of chronic, immune-mediated colitis. Front Immunol. 2019;10:1420. doi: 10.3389/fimmu.2019.01420.
44. Palmela C, Chevarin C, Xu Z, Torres J, Sevrin G, Hirten R, Barnich N, Ng SC, Colombel J-F. Adherent-invasive *Escherichia coli* in inflammatory bowel disease. Gut. 2018;67(3):574–587. doi: 10.1136/gutjnl-2017-314903.
45. Kullberg MC, Jankovic D, Feng CG, Hue S, Gorelick PL, McKenzie BS, Cua DJ, Powrie F, Cheever AW, Maloy KJ, et al. IL-23 plays a key role in helicobacter hepaticus-induced T cell-dependent colitis. J Experiment Med. 2006;203(11):2485–2494. doi: 10.1084/jem.20061082.
46. Anderson MS, Venanzi ES, Klein L, Chen Z, Berzins SP, Turley SJ, von Boehmer H, Bronson R, Dierich A, Benoist C, et al. Projection of an immunological self shadow within the thymus by the aire protein. Science. 2002;298(5597):1395–1401. doi: 10.1126/science.1075958.
47. Liston A, Lesage S, Wilson J, Peltonen L, Goodnow CC. Aire regulates negative selection of organ-specific T cells. Nat Immunol. 2003;4(4):350–354. doi: 10.1038/ni906.
48. Takaba H, Morishita Y, Tomofuji Y, Danks L, Nitta T, Komatsu N, Kodama T, Takayanagi H. Fezf2 orchestrates a thymic program of self-antigen expression for immune tolerance. Cell. 2015;163(4):975–987. doi: 10.1016/j.cell.2015.10.013.
49. Voboril M, Brabec T, Dobes J, Splichalova I, Brezina J, Cepkova A, Dobešová M, Aidarova A, Kubovčiak J, Tsyklauri O, et al. Toll-like receptor signaling in thymic epithelium controls monocyte-derived dendritic cell recruitment and Treg generation. Nat Commun. 2020;11(1):2361. doi: 10.1038/s41467-020-16081-3.
50. Leventhal DS, Gilmore DC, Berger JM, Nishi S, Lee V, Malchow S, Kline D, Kline J, Vander Griend D, Huang H, et al. Dendritic cells coordinate the development and homeostasis of organ-specific regulatory T cells. Immunity. 2016;44(4):847–859. doi: 10.1016/j.immuni.2016.01.025.
51. Perry JSA, Lio C-W, Kau AL, Nutsch K, Yang Z, Gordon JI, Murphy K, Hsieh C-S. Distinct contributions of aire and antigen-presenting-cell subsets to the generation of self-tolerance in the thymus. Immunity. 2014;41(3):414–426. doi: 10.1016/j.immuni.2014.08.007.
52. Zegarra-Ruiz DF, Kim DV, Norwood K, Kim M, Wu WH, Saldana-Morales FB, Hill AA, Majumdar S, Orozco S, Bell R, et al. Thymic development of gut-

microbiota-specific T cells. Nature. 2021;594 (7863):413–417. doi: 10.1038/s41586-021-03531-1.
53. Byun S, Lee J, Choi YH, Ko H, Lee C, Park JC, Kim SW, Lee H, Sharma A, Kim KS, et al. Gut microbiota defines functional direction of colonic regulatory T cells with unique TCR repertoires. J Immunol. 2024;213(6):886–897. doi: 10.4049/jimmunol.2300395.
54. Blanco T, Singh RB, Nakagawa H, Taketani Y, Dohlman TH, Chen Y, Chauhan SK, Yin J, Dana R. Conventional type I migratory CD103+ dendritic cells are required for corneal allograft survival. Mucosal Immunol. 2023;16(5):711–726. doi: 10.1016/j.mucimm.2022.12.002.
55. Curotto de Lafaille MA, Kutchukhidze N, Shen S, Ding Y, Yee H, Lafaille JJ. Adaptive Foxp3+ regulatory T cell-dependent and -independent control of allergic inflammation. Immunity. 2008;29(1):114–126. doi: 10.1016/j.immuni.2008.05.010.
56. Esterhazy D, Loschko J, London M, Jove V, Oliveira TY, Mucida D. Classical dendritic cells are required for dietary antigen-mediated induction of peripheral T (reg) cells and tolerance. Nat Immunol. 2016;17 (5):545–555. doi: 10.1038/ni.3408.
57. Gribonika I, Stromberg A, Chandode RK, Schon K, Lahl K, Bemark M, Lycke N. Migratory CD103(+) CD11b(+) cDc2s in Peyer's patches are critical for gut IgA responses following oral immunization. Mucosal Immunol. 2024;17(4):509–523. doi: 10.1016/j.mucimm.2024.03.004.
58. Iberg CA, Hawiger D. Natural and induced tolerogenic dendritic cells. The J Immunol. 2020;204(4):733–744. doi: 10.4049/jimmunol.1901121.
59. Josefowicz SZ, Niec RE, Kim HY, Treuting P, Chinen T, Zheng Y, Umetsu DT, Rudensky AY. Extrathymically generated regulatory T cells control mucosal TH2 inflammation. Nature. 2012;482(7385):395–399. doi: 10.1038/nature10772.
60. Russler-Germain EV, Yi J, Young S, Nutsch K, Wong HS, Ai TL, Chai JN, Durai V, Kaplan DH, Germain RN, et al. Gut Helicobacter presentation by multiple dendritic cell subsets enables context-specific regulatory T cell generation. Elife. 2021;10:10. doi: 10.7554/eLife.54792.
61. Akagbosu B, Tayyebi Z, Shibu G, Paucar Iza YA, Deep D, Parisotto YF, Fisher L, Pasolli HA, Thevin V, Elmentaite R, et al. Novel antigen-presenting cell imparts Treg-dependent tolerance to gut microbiota. Nature. 2022;610(7933):752–760. doi: 10.1038/s41586-022-05309-5.
62. Kedmi R, Najar TA, Mesa KR, Grayson A, Kroehling L, Hao Y, Hao S, Pokrovskii M, Xu M, Talbot J, et al. A RORγt+ cell instructs gut microbiota-specific Treg cell differentiation. Nature. 2022;610(7933):737–743. doi: 10.1038/s41586-022-05089-y.
63. Lyu M, Suzuki H, Kang L, Gaspal F, Zhou W, Goc J, Zhou L, Zhou J, Zhang W, Artis D, et al. ILC3s select microbiota-specific regulatory T cells to establish tolerance in the gut. Nature. 2022;610(7933):744–751. doi: 10.1038/s41586-022-05141-x.
64. Elmentaite R, Kumasaka N, Roberts K, Fleming A, Dann E, King HW, Kleshchevnikov V, Dabrowska M, Pritchard S, Bolt L, et al. Cells of the human intestinal tract mapped across space and time. Nature. 2021;597 (7875):250–255. doi: 10.1038/s41586-021-03852-1.
65. Vatanen T, Kostic AD, d'Hennezel E, Siljander H, Franzosa EA, Yassour M, Kolde R, Vlamakis H, Arthur TD, Hämäläinen A-M, et al. Variation in microbiome LPS immunogenicity contributes to autoimmunity in humans. Cell. 2016;165(6):1551. doi: 10.1016/j.cell.2016.05.056.
66. Sollid LM, Jabri B. Triggers and drivers of autoimmunity: lessons from coeliac disease. Nat Rev Immunol. 2013;13(4):294–302. doi: 10.1038/nri3407.
67. Kubinak JL, Stephens WZ, Soto R, Petersen C, Chiaro T, Gogokhia L, Bell R, Ajami NJ, Petrosino JF, Morrison L, et al. MHC variation sculpts individualized microbial communities that control susceptibility to enteric infection. Nat Commun. 2015;6(1):8642. doi: 10.1038/ncomms9642.
68. Royet J, Gupta D, Dziarski R. Peptidoglycan recognition proteins: modulators of the microbiome and inflammation. Nat Rev Immunol. 2011;11(12):837–851. doi: 10.1038/nri3089.
69. Brown S, Santa Maria JP, Walker S. Wall teichoic acids of gram-positive bacteria. Annu Rev Microbiol. 2013;67 (1):313–336. doi: 10.1146/annurev-micro-092412-155620.
70. Kwon HK, Lee CG, So JS, Chae CS, Hwang JS, Sahoo A, Nam JH, Rhee JH, Hwang K-C, Im S-H. Generation of regulatory dendritic cells and CD4+Foxp3+ T cells by probiotics administration suppresses immune disorders. Proc Natl Acad Sci USA. 2010;107(5):2159–2164. doi: 10.1073/pnas.0904055107.
71. Kim JE, Sharma A, Sharma G, Lee SY, Shin HS, Rudra D, Im S-H. Lactobacillus pentosus modulates immune response by inducing IL-10 producing Tr1 cells. Immune Netw. 2019;19(6):e39. doi: 10.4110/in.2019.19.e39.
72. Garcia-Vello P, Sharma G, Speciale I, Molinaro A, Im SH, De Castro C. Structural features and immunological perception of the cell surface glycans of lactobacillus plantarum: a novel rhamnose-rich polysaccharide and teichoic acids. Carbohydr Polym. 2020;233:115857. doi: 10.1016/j.carbpol.2020.115857.
73. Sharma G, Sharma A, Kim I, Cha DG, Kim S, Park ES, Noh JG, Lee J, Ku JH, Choi YH, et al. A dietary commensal microbe enhances antitumor immunity by activating tumor macrophages to sequester iron. Nat Immunol. 2024;25(5):790–801. doi: 10.1038/s41590-024-01816-x.
74. Round JL, Lee SM, Li J, Tran G, Jabri B, Chatila TA, Mazmanian SK. The toll-like receptor 2 pathway establishes colonization by a commensal of the human

microbiota. Science. 2011;332(6032):974–977. doi: 10.1126/science.1206095.
75. Kreisman LS, Cobb BA. Glycoantigens induce human peripheral Tr1 cell differentiation with gut-homing specialization. J Biol Chem. 2011;286(11):8810–8818. doi: 10.1074/jbc.M110.206011.
76. Telesford KM, Yan W, Ochoa-Reparaz J, Pant A, Kircher C, Christy MA, Begum-Haque S, Kasper DL, Kasper LH. A commensal symbiotic factor derived from bacteroides fragilis promotes human CD39 + Foxp3 + T cells and T reg function. Gut Microbes. 2015;6(4):234–242. doi: 10.1080/19490976.2015.1056973.
77. Carasso S, Zaatry R, Hajjo H, Kadosh-Kariti D, Ben-Assa N, Naddaf R, Mandelbaum N, Pressman S, Chowers Y, Gefen T, et al. Inflammation and bacteriophages affect DNA inversion states and functionality of the gut microbiota. Cell Host & Microbe. 2024;32(3):322–334.e9. doi: 10.1016/j.chom.2024.02.003.
78. Blandford LE, Johnston EL, Sanderson JD, Wade WG, Lax AJ. Promoter orientation of the immunomodulatory bacteroides fragilis capsular polysaccharide a (PSA) is off in individuals with inflammatory bowel disease (IBD). Gut Microbes. 2019;10(5):569–577. doi: 10.1080/19490976.2018.1560755.
79. Neff CP, Rhodes ME, Arnolds KL, Collins CB, Donnelly J, Nusbacher N, Jedlicka P, Schneider J, McCarter M, Shaffer M, et al. Diverse intestinal bacteria contain putative zwitterionic capsular polysaccharides with anti-inflammatory properties. Cell Host & Microbe. 2016;20(4):535–547. doi: 10.1016/j.chom.2016.09.002.
80. Turroni F, Foroni E, Pizzetti P, Giubellini V, Ribbera A, Merusi P, Cagnasso P, Bizzarri B, de'Angelis GL, Shanahan F, et al. Exploring the diversity of the bifidobacterial population in the human intestinal tract. Appl Environ Microbiol. 2009;75(6):1534–1545. doi: 10.1128/AEM.02216-08.
81. Kirjavainen PV, Arvola T, Salminen SJ, Isolauri E. Aberrant composition of gut microbiota of allergic infants: a target of bifidobacterial therapy at weaning? Gut. 2002;51(1):51–55. doi: 10.1136/gut.51.1.51.
82. Speciale I, Verma R, Di Lorenzo F, Molinaro A, Im SH, De Castro C. Bifidobacterium bifidum presents on the cell surface a complex mixture of glucans and galactans with different immunological properties. Carbohydr Polym. 2019;218:269–278. doi: 10.1016/j.carbpol.2019.05.006.
83. Nyirenda MH, Sanvito L, Darlington PJ, O'Brien K, Zhang GX, Constantinescu CS, Bar-Or A, Gran B. TLR2 stimulation drives human naive and effector regulatory T cells into a Th17-like phenotype with reduced suppressive function. J Of Immunol. 2011;187(5):2278–2290. doi: 10.4049/jimmunol.1003715.
84. Oberg HH, Juricke M, Kabelitz D, Wesch D. Regulation of T cell activation by TLR ligands. Eur J Cell Biol. 2011;90(6–7):582–592. doi: 10.1016/j.ejcb.2010.11.012.
85. Doron I, Leonardi I, Li XV, Fiers WD, Semon A, Bialt-DeCelie M, Migaud M, Gao IH, Lin W-Y, Kusakabe T, et al. Human gut mycobiota tune immunity via CARD9-dependent induction of anti-fungal IgG antibodies. Cell. 2021;184(4):1017–1031.e14. doi: 10.1016/j.cell.2021.01.016.
86. Chehoud C, Albenberg LG, Judge C, Hoffmann C, Grunberg S, Bittinger K, Baldassano RN, Lewis JD, Bushman FD, Wu GD. Fungal signature in the gut microbiota of pediatric patients with inflammatory bowel disease. Inflammat Bowel Dis. 2015;21(8):1948–1956. doi: 10.1097/MIB.0000000000000454.
87. Underhill DM, Iliev ID. The mycobiota: interactions between commensal fungi and the host immune system. Nat Rev Immunol. 2014;14(6):405–416. doi: 10.1038/nri3684.
88. Hoarau G, Mukherjee PK, Gower-Rousseau C, Hager C, Chandra J, Retuerto MA, Neut C, Vermeire S, Clemente J, Colombel JF, et al. Bacteriome and mycobiome interactions underscore microbial dysbiosis in familial Crohn's disease. mBio. 2016;7(5). doi: 10.1128/mBio.01250-16.
89. Liguori G, Lamas B, Richard ML, Brandi G, da Costa G, Hoffmann TW, Di Simone MP, Calabrese C, Poggioli G, Langella P, et al. Fungal dysbiosis in mucosa-associated microbiota of Crohn's disease patients. J Crohn's Colitis. 2016;10(3):296–305. doi: 10.1093/ecco-jcc/jjv209.
90. Jain U, Ver Heul AM, Xiong S, Gregory MH, Demers EG, Kern JT, Lai C-W, Muegge BD, Barisas DAG, Leal-Ekman JS, et al. Debaryomyces is enriched in Crohn's disease intestinal tissue and impairs healing in mice. Science. 2021;371(6534):1154–1159. doi: 10.1126/science.abd0919.
91. Israeli E, Grotto I, Gilburd B, Balicer RD, Goldin E, Wiik A. Anti-saccharomyces cerevisiae and antineutrophil cytoplasmic antibodies as predictors of inflammatory bowel disease. Gut. 2005;54(9):1232–1236. doi: 10.1136/gut.2004.060228.
92. Odds FC, Davidson AD, Jacobsen MD, Tavanti A, Whyte JA, Kibbler CC, Ellis DH, Maiden MCJ, Shaw DJ, Gow NAR. Candida albicans strain maintenance, replacement, and microvariation demonstrated by multilocus sequence typing. J Clin Microbiol. 2006;44(10):3647–3658. doi: 10.1128/JCM.00934-06.
93. Standaert–Vitse A, Jouault T, Vandewalle P, Mille C, Seddik M, Sendid B, Mallet J, Colombel J, Poulain D. Candida albicans is an immunogen for anti–saccharomyces cerevisiae antibody markers of Crohn's disease. Gastroenterol. 2006;130(6):1764–1775. doi: 10.1053/j.gastro.2006.02.009.
94. Novak M, Vetvicka V. β -glucans, history, and the present: immunomodulatory aspects and mechanisms of action. J Of Immunotoxicol. 2008;5(1):47–57. doi: 10.1080/15476910802019045.

95. Reid DM, Gow NA, Brown GD. Pattern recognition: recent insights from Dectin-1. Curr Opin Immunol. 2009;21(1):30–37. doi: 10.1016/j.coi.2009.01.003.
96. Koh A, De Vadder F, Kovatcheva-Datchary P, Backhed F. From dietary fiber to host physiology: short-chain fatty acids as key bacterial metabolites. Cell. 2016;165(6):1332–1345. doi: 10.1016/j.cell.2016.05.041.
97. Ratajczak W, Ryl A, Mizerski A, Walczakiewicz K, Sipak O, Laszczynska M. Immunomodulatory potential of gut microbiome-derived short-chain fatty acids (SCFAs). Acta Biochim Pol. 2019;66(1):1–12. doi: 10.18388/abp.2018_2648.
98. Marino E, Richards JL, McLeod KH, Stanley D, Yap YA, Knight J, McKenzie C, Kranich J, Oliveira AC, Rossello FJ, et al. Gut microbial metabolites limit the frequency of autoimmune T cells and protect against type 1 diabetes. Nat Immunol. 2017;18(5):552–562. doi: 10.1038/ni.3713.
99. Smith PM, Howitt MR, Panikov N, Michaud M, Gallini CA, Bohlooly YM, Glickman JN, Garrett WS. The microbial metabolites, short-chain fatty acids, regulate colonic Treg cell homeostasis. Science. 2013;341(6145):569–573. doi: 10.1126/science.1241165.
100. Zhuge A, Li S, Han S, Yuan Y, Shen J, Wu W, Wang K, Xia J, Wang Q, Gu Y, et al. Akkermansia muciniphila-derived acetate activates the hepatic AMPK/SIRT1/PGC-1α axis to alleviate ferroptosis in metabolic-associated fatty liver disease. Acta Pharmaceutica Sinica B. 2025;15(1):151–167. doi: 10.1016/j.apsb.2024.10.010.
101. Lakshmanan AP, Murugesan S, Al Khodor S, Terranegra A. The potential impact of a probiotic: akkermansia muciniphila in the regulation of blood pressure—the current facts and evidence. J Transl Med. 2022;20(1):430. doi: 10.1186/s12967-022-03631-0.
102. Fukuda S, Toh H, Hase K, Oshima K, Nakanishi Y, Yoshimura K, Tobe T, Clarke JM, Topping DL, Suzuki T, et al. Bifidobacteria can protect from enteropathogenic infection through production of acetate. Nature. 2011;469(7331):543–547. doi: 10.1038/nature09646.
103. Yoon SJ, Yu JS, Min BH, Gupta H, Won SM, Park HJ, Han SH, Kim B-Y, Kim KH, Kim BK, et al. Bifidobacterium-derived short-chain fatty acids and indole compounds attenuate nonalcoholic fatty liver disease by modulating gut-liver axis. Front Microbiol. 2023;14:1129904. doi: 10.3389/fmicb.2023.1129904.
104. O'Riordan KJ, Collins MK, Moloney GM, Knox EG, Aburto MR, Fulling C, Morley SJ, Clarke G, Schellekens H, Cryan JF. Short chain fatty acids: microbial metabolites for gut-brain axis signalling. Molecular and Cellular Endocrinol. 2022;546:111572. doi: 10.1016/j.mce.2022.111572.
105. Rios-Covian D, Gueimonde M, Duncan SH, Flint HJ, de Los reyes-Gavilan CG, Cox M. Enhanced butyrate formation by cross-feeding between faecalibacterium prausnitzii and bifidobacterium adolescentis. FEMS Microbiol Lett. 2015;362(21):fnv176. doi: 10.1093/femsle/fnv176.
106. Martin R, Rios-Covian D, Huillet E, Auger S, Khazaal S, Bermudez-Humaran LG, Sokol H, Chatel J-M, Langella P. Faecalibacterium: a bacterial genus with promising human health applications. FEMS Microbiol Rev. 2023;47(4). doi: 10.1093/femsre/fuad039.
107. van der Lelie D, Oka A, Taghavi S, Umeno J, Fan TJ, Merrell KE, Watson SD, Ouellette L, Liu B, Awoniyi M, et al. Rationally designed bacterial consortia to treat chronic immune-mediated colitis and restore intestinal homeostasis. Nat Commun. 2021;12(1):3105. doi: 10.1038/s41467-021-23460-x.
108. Wang X, Cai Z, Wang Q, Wu C, Sun Y, Wang Z, Xu X, Xue W, Cao Z, Zhang M, et al. Bacteroides methylmalonyl-CoA mutase produces propionate that promotes intestinal goblet cell differentiation and homeostasis. Cell Host & Microbe. 2024;32(1):63–78.e7. doi: 10.1016/j.chom.2023.11.005.
109. Zhou Y, Xu H, Xu J, Guo X, Zhao H, Chen Y, Zhou Y, Nie Y. F. prausnitzii and its supernatant increase SCFAs-producing bacteria to restore gut dysbiosis in TNBS-induced colitis. AMB Express. 2021;11(1):33. doi: 10.1186/s13568-021-01197-6.
110. Mukherjee A, Lordan C, Ross RP, Cotter PD. Gut microbes from the phylogenetically diverse genus eubacterium and their various contributions to gut health. Gut Microbes. 2020;12(1):1802866. doi: 10.1080/19490976.2020.1802866.
111. Lu H, Xu X, Fu D, Gu Y, Fan R, Yi H, He X, Wang C, Ouyang B, Zhao P, et al. Butyrate-producing eubacterium rectale suppresses lymphomagenesis by alleviating the TNF-induced TLR4/MyD88/NF-kappaB axis. Cell Host & Microbe. 2022;30(8):1139–1150.e7. doi: 10.1016/j.chom.2022.07.003.
112. Yano JM, Yu K, Donaldson GP, Shastri GG, Ann P, Ma L, Nagler C, Ismagilov R, Mazmanian S, Hsiao E. Indigenous bacteria from the gut microbiota regulate host serotonin biosynthesis. Cell. 2015;161(2):264–276. doi: 10.1016/j.cell.2015.02.047.
113. O'Mahony SM, Clarke G, Borre YE, Dinan TG, Cryan JF. SerotonIn, tryptophan metabolism and the brain-gut-microbiome axis. Behav Brain Res. 2015;277:32–48. doi: 10.1016/j.bbr.2014.07.027.
114. Desbonnet L, Garrett L, Clarke G, Bienenstock J, Dinan TG. The probiotic bifidobacteria infantis: an assessment of potential antidepressant properties in the rat. J Psychiatric Res. 2008;43(2):164–174. doi: 10.1016/j.jpsychires.2008.03.009.
115. Wikoff WR, Anfora AT, Liu J, Schultz PG, Lesley SA, Peters EC, Siuzdak G. Metabolomics analysis reveals large effects of gut microflora on mammalian blood metabolites. Proc Natl Acad Sci USA. 2009;106(10):3698–3703. doi: 10.1073/pnas.0812874106.
116. Reigstad CS, Salmonson CE, Iii JF, Szurszewski JH, Linden DR, Sonnenburg JL, Farrugia G, Kashyap PC. Gut microbes promote colonic serotonin production

through an effect of short-chain fatty acids on enterochromaffin cells. FASEB J. 2015;29(4):1395–1403. doi: 10.1096/fj.14-259598.
117. Fukumoto S, Tatewaki M, Yamada T, Fujimiya M, Mantyh C, Voss M, Eubanks S, Harris M, Pappas TN, Takahashi T. Short-chain fatty acids stimulate colonic transit via intraluminal 5-HT release in rats. Am J Of Physiol-Regulat, Integrat Comparat Physiol. 2003;284(5):R1269–76. doi: 10.1152/ajpregu.00442.2002.
118. Lyte M. Probiotics function mechanistically as delivery vehicles for neuroactive compounds: microbial endocrinology in the design and use of probiotics. Bioessays. 2011;33(8):574–581. doi: 10.1002/bies.201100024.
119. Nohr MK, Pedersen MH, Gille A, Egerod KL, Engelstoft MS, Husted AS, Sichlau RM, Grunddal KV, Seier Poulsen S, Han S, et al. GPR41/FFAR3 and GPR43/FFAR2 as cosensors for short-chain fatty acids in enteroendocrine cells vs FFAR3 in enteric neurons and FFAR2 in enteric leukocytes. Endocrinol. 2013;154(10):3552–3564. doi: 10.1210/en.2013-1142.
120. Pluznick JL. Microbial short-chain fatty acids and blood pressure regulation. Curr Hypertens Rep. 2017;19(4):25. doi: 10.1007/s11906-017-0722-5.
121. Thangaraju M, Cresci GA, Liu K, Ananth S, Gnanaprakasam JP, Browning DD, Mellinger JD, Smith SB, Digby GJ, Lambert NA, et al. GPR109A is a G-protein–coupled receptor for the bacterial fermentation product butyrate and functions as a tumor suppressor in colon. Cancer Res. 2009;69(7):2826–2832. doi: 10.1158/0008-5472.CAN-08-4466.
122. Le Poul E, Loison C, Struyf S, Springael JY, Lannoy V, Decobecq ME, Brezillon S, Dupriez V, Vassart G, Van Damme J, et al. Functional characterization of human receptors for short chain fatty acids and their role in polymorphonuclear cell activation. J Of Biolog Chem. 2003;278(28):25481–25489. doi: 10.1074/jbc.M301403200.
123. Taggart AK, Kero J, Gan X, Cai TQ, Cheng K, Ippolito M, Ren N, Kaplan R, Wu K, Wu T-J, et al. (d)-β-hydroxybutyrate inhibits adipocyte lipolysis via the nicotinic acid receptor PUMA-G. J Of Biol Chem. 2005;280(29):26649–26652. doi: 10.1074/jbc.C500213200.
124. Gerriets VA, Kishton RJ, Nichols AG, Macintyre AN, Inoue M, Ilkayeva O, Winter PS, Liu X, Priyadharshini B, Slawinska ME, et al. Metabolic programming and PDHK1 control CD4+ T cell subsets and inflammation. J Clin Invest. 2015;125(1):194–207. doi: 10.1172/JCI76012.
125. Kimura I, Inoue D, Maeda T, Hara T, Ichimura A, Miyauchi S, Kobayashi M, Hirasawa A, Tsujimoto G. Short-chain fatty acids and ketones directly regulate sympathetic nervous system via G protein-coupled receptor 41 (GPR41). Proc Natl Acad Sci USA. 2011;108(19):8030–8035. doi: 10.1073/pnas.1016088108.
126. Singh N, Gurav A, Sivaprakasam S, Brady E, Padia R, Shi H, Thangaraju M, Prasad P, Manicassamy S, Munn D, et al. Activation of Gpr109a, receptor for niacin and the commensal metabolite butyrate, suppresses colonic inflammation and carcinogenesis. Immunity. 2014;40(1):128–139. doi: 10.1016/j.immuni.2013.12.007.
127. Macia L, Tan J, Vieira AT, Leach K, Stanley D, Luong S, Maruya M, Ian mckenzie C, Hijikata A, Wong C, et al. Metabolite-sensing receptors GPR43 and GPR109A facilitate dietary fibre-induced gut homeostasis through regulation of the inflammasome. Nat Commun. 2015;6(1):6734. doi: 10.1038/ncomms7734.
128. Park J, Kim M, Kang SG, Jannasch AH, Cooper B, Patterson J, Kim CH. Short-chain fatty acids induce both effector and regulatory T cells by suppression of histone deacetylases and regulation of the mTOR–S6K pathway. Mucosal Immunol. 2015;8(1):80–93. doi: 10.1038/mi.2014.44.
129. Duscha A, Gisevius B, Hirschberg S, Yissachar N, Stangl GI, Dawin E, Bader V, Haase S, Kaisler J, David C, et al. Propionic acid shapes the multiple sclerosis disease course by an immunomodulatory mechanism. Cell. 2020;180(6):1067–1080.e16. doi: 10.1016/j.cell.2020.02.035.
130. Huda-Faujan N, Abdulamir AS, Fatimah AB, Anas OM, Shuhaimi M, Yazid AM, Loong YY. The impact of the level of the intestinal short chain fatty acids in inflammatory bowel disease patients versus healthy subjects. Open Biochem J. 2010;4:53–58. doi: 10.2174/1874091X01004010053.
131. Agus A, Clement K, Sokol H. Gut microbiota-derived metabolites as central regulators in metabolic disorders. Gut. 2021;70(6):1174–1182. doi: 10.1136/gutjnl-2020-323071.
132. Parada Venegas D, De la Fuente MK, Landskron G, Gonzalez MJ, Quera R, Dijkstra G, Harmsen HJM, Faber KN, Hermoso MA. Short chain fatty acids (SCFAs)-mediated gut epithelial and immune regulation and its relevance for inflammatory bowel diseases. Front Immunol. 2019;10:277. doi: 10.3389/fimmu.2019.00277.
133. Machiels K, Joossens M, Sabino J, De Preter V, Arijs I, Eeckhaut V, Ballet V, Claes K, Van Immerseel F, Verbeke K, et al. A decrease of the butyrate-producing species roseburia hominis and faecalibacterium prausnitzii defines dysbiosis in patients with ulcerative colitis. Gut. 2014;63(8):1275–1283. doi: 10.1136/gutjnl-2013-304833.
134. Park J, Wang Q, Wu Q, Mao-Draayer Y, Kim CH. Bidirectional regulatory potentials of short-chain fatty acids and their G-protein-coupled receptors in autoimmune neuroinflammation. Sci Rep. 2019;9(1):8837. doi: 10.1038/s41598-019-45311-y.
135. de Goffau MC, Fuentes S, van den Bogert B, Honkanen H, de Vos WM, Welling GW, Hyöty H, Harmsen HJM. Aberrant gut microbiota composition at the onset of

type 1 diabetes in young children. Diabetologia. 2014;57(8):1569–1577. doi: 10.1007/s00125-014-3274-0.
136. Arrieta MC, Stiemsma LT, Dimitriu PA, Thorson L, Russell S, Yurist-Doutsch S, Kuzeljevic B, Gold MJ, Britton HM, Lefebvre DL, et al. Early infancy microbial and metabolic alterations affect risk of childhood asthma. Sci Transl Med. 2015;7(307):307ra152. doi: 10.1126/scitranslmed.aab2271.
137. Brown AJ, Goldsworthy SM, Barnes AA, Eilert MM, Tcheang L, Daniels D, Muir AI, Wigglesworth MJ, Kinghorn I, Fraser NJ, et al. The orphan G protein-coupled receptors GPR41 and GPR43 are activated by propionate and other short chain carboxylic acids. J Of BiolChem. 2003;278(13):11312–11319. doi: 10.1074/jbc.M211609200.
138. Ang Z, Ding JL. GPR41 and GPR43 in obesity and inflammation - protective or causative? Front Immunol. 2016;7:28. doi: 10.3389/fimmu.2016.00028.
139. Correa-Oliveira R, Fachi JL, Vieira A, Sato FT, Vinolo MA. Regulation of immune cell function by short-chain fatty acids. Clin & Trans Imm. 2016;5(4):e73. doi: 10.1038/cti.2016.17.
140. Kespohl M, Vachharajani N, Luu M, Harb H, Pautz S, Wolff S, Sillner N, Walker A, Schmitt-Kopplin P, Boettger T, et al. The microbial metabolite butyrate induces expression of Th1-associated factors in CD4 (+) T cells. Front Immunol. 2017;8:1036. doi: 10.3389/fimmu.2017.01036.
141. Miller KD, O'Connor S, Pniewski KA, Kannan T, Acosta R, Mirji G, Papp S, Hulse M, Mukha D, Hlavaty SI, et al. Acetate acts as a metabolic immunomodulator by bolstering T-cell effector function and potentiating antitumor immunity in breast cancer. Nat Cancer. 2023;4(10):1491–1507. doi: 10.1038/s43018-023-00636-6.
142. Qiu J, Villa M, Sanin DE, Buck MD, O'Sullivan D, Ching R, Matsushita M, Grzes KM, Winkler F, Chang C-H, et al. Acetate promotes T cell effector function during glucose restriction. Cell Reports. 2019;27(7):2063–2074.e5. doi: 10.1016/j.celrep.2019.04.022.
143. Balmer ML, Ma EH, Bantug GR, Grahlert J, Pfister S, Glatter T, Jauch A, Dimeloe S, Slack E, Dehio P, et al. Memory CD8 + T cells require increased concentrations of acetate induced by stress for optimal function. Immunity. 2016;44(6):1312–1324. doi: 10.1016/j.immuni.2016.03.016.
144. Hague A, Manning AM, Hanlon KA, Huschtscha LI, Hart D, Paraskeva C. Sodium butyrate induces apoptosis in human colonic tumour cell lines in a p53-independent pathway: implications for the possible role of dietary fibre in the prevention of large-bowel cancer. Intl Journal Of Cancer. 1993;55(3):498–505. doi: 10.1002/ijc.2910550329.
145. Shimizu T, Ohtake H, Fujii T, Tabuchi Y, Sakai H. Volume-sensitive outwardly rectifying Cl– channels contribute to butyrate-triggered apoptosis of murine colonic epithelial MCE301 cells. Journal Of Physiol Sciences. 2015;65(2):151–157. doi: 10.1007/s12576-014-0352-5.
146. Donohoe DR, Holley D, Collins LB, Montgomery SA, Whitmore AC, Hillhouse A, Curry KP, Renner SW, Greenwalt A, Ryan EP, et al. A gnotobiotic mouse model demonstrates that dietary fiber protects against colorectal tumorigenesis in a microbiota- and butyrate-dependent manner. Cancer Discovery. 2014;4(12):1387–1397. doi: 10.1158/2159-8290.CD-14-0501.
147. Nastasi C, Fredholm S, Willerslev-Olsen A, Hansen M, Bonefeld CM, Geisler C, Andersen MH, Ødum N, Woetmann A. Butyrate and propionate inhibit antigen-specific CD8(+) T cell activation by suppressing IL-12 production by antigen-presenting cells. Sci Rep. 2017;7(1):14516. doi: 10.1038/s41598-017-15099-w.
148. Erny D, Hrabe de Angelis AL, Jaitin D, Wieghofer P, Staszewski O, David E, Keren-Shaul H, Mahlakoiv T, Jakobshagen K, Buch T, et al. Host microbiota constantly control maturation and function of microglia in the CNS. Nat Neurosci. 2015;18(7):965–977. doi: 10.1038/nn.4030.
149. Mizuno M, Noto D, Kaga N, Chiba A, Miyake S, Ashour HM. The dual role of short fatty acid chains in the pathogenesis of autoimmune disease models. PLOS ONE. 2017;12(2):e0173032. doi: 10.1371/journal.pone.0173032.
150. Zeng X, Xing X, Gupta M, Keber FC, Lopez JG, Lee YCJ, Roichman A, Wang L, Neinast MD, Donia MS, et al. Gut bacterial nutrient preferences quantified in vivo. Cell. 2022;185(18):3441–3456.e19. doi: 10.1016/j.cell.2022.07.020.
151. Alkhalaf LM, Ryan KS. Biosynthetic manipulation of tryptophan in bacteria: pathways and mechanisms. Chem Biol. 2015;22(3):317–328. doi: 10.1016/j.chembiol.2015.02.005.
152. Mellor AL, Munn DH. IDO expression by dendritic cells: tolerance and tryptophan catabolism. Nat Rev Immunol. 2004;4(10):762–774. doi: 10.1038/nri1457.
153. Platten M, Wick W, Van den Eynde BJ. Tryptophan catabolism in cancer: beyond IDO and tryptophan depletion. Cancer Res. 2012;72(21):5435–5440. doi: 10.1158/0008-5472.CAN-12-0569.
154. Stone TW, Darlington LG. The kynurenine pathway as a therapeutic target in cognitive and neurodegenerative disorders. Br J Pharmacol. 2013;169(6):1211–1227. doi: 10.1111/bph.12230.
155. Gershon MD, Tack J. The serotonin signaling system: from basic understanding to drug development for functional GI disorders. Gastroenterology. 2007;132(1):397–414. doi: 10.1053/j.gastro.2006.11.002.
156. Martin CR, Osadchiy V, Kalani A, Mayer EA. The brain-gut-microbiome axis. Cellular Mol Gastroenterol Hepatol. 2018;6(2):133–148. doi: 10.1016/j.jcmgh.2018.04.003.
157. Taleb S. Tryptophan dietary impacts gut barrier and metabolic diseases. Front Immunol. 2019;10:2113. doi: 10.3389/fimmu.2019.02113.

158. Roager HM, Hansen LB, Bahl MI, Frandsen HL, Carvalho V, Gobel RJ, Dalgaard MD, Plichta DR, Sparholt MH, Vestergaard H, et al. Colonic transit time is related to bacterial metabolism and mucosal turnover in the gut. Nat Microbiol. 2016;1(9):16093. doi: 10.1038/nmicrobiol.2016.93.
159. Zelante T, Iannitti RG, Cunha C, De Luca A, Giovannini G, Pieraccini G, Zecchi R, D'Angelo C, Massi-Benedetti C, Fallarino F, et al. Tryptophan catabolites from microbiota engage aryl hydrocarbon receptor and balance mucosal reactivity via interleukin-22. Immunity. 2013;39(2):372–385. doi: 10.1016/j.immuni.2013.08.003.
160. Wu H, Herr D, MacIver NJ, Rathmell JC, Gerriets VA. CD4 T cells differentially express cellular machinery for serotonin signaling, synthesis, and metabolism. Int Immunopharmacol. 2020;88:106922. doi: 10.1016/j.intimp.2020.106922.
161. Dinan TG, Stilling RM, Stanton C, Cryan JF. Collective unconscious: how gut microbes shape human behavior. J Psychiatric Res. 2015;63:1–9. doi: 10.1016/j.jpsychires.2015.02.021.
162. Özoğul F. Production of biogenic amines by morganella morganii, Klebsiella pneumoniae and hafnia alvei using a rapid HPLC method. Eur Food Res Technol. 2004;219(5):465–469. doi: 10.1007/s00217-004-0988-0.
163. Shishov VA, Kirovskaia TA, Kudrin VS, Oleskin AV. Amine neuromediators, their precursors, and oxidation products in the culture of *Escherichia coli* K-12. Appl Biochem Microbiol. 2009;45(5):494–497. doi: 10.1134/S0003683809050068.
164. Smith EA, Macfarlane GT. Enumeration of human colonic bacteria producing phenolic and indolic compounds: effects of pH, carbohydrate availability and retention time on dissimilatory aromatic amino acid metabolism. J Appl Bacteriol. 1996;81(3):288–302. doi: 10.1111/j.1365-2672.1996.tb04331.x.
165. Lee JH, Lee J. Indole as an intercellular signal in microbial communities. FEMS Microbiol Rev. 2010;34(4):426–444. doi: 10.1111/j.1574-6976.2009.00204.x.
166. Buffie CG, Bucci V, Stein RR, McKenney PT, Ling L, Gobourne A, No D, Liu H, Kinnebrew M, Viale A, et al. Precision microbiome reconstitution restores bile acid mediated resistance to clostridium difficile. Nature. 2015;517(7533):205–208. doi: 10.1038/nature13828.
167. Chen C, Ye Y, Wang R, Zhang Y, Wu C, Debnath SC, Ma Z, Wang J, Wu M. Streptomyces nigra sp. nov. Is a novel actinobacterium isolated from mangrove soil and exerts a potent antitumor activity in vitro. Front Microbiol. 2018;9:1587. doi: 10.3389/fmicb.2018.01587.
168. Elsden SR, Hilton MG, Waller JM. The end products of the metabolism of aromatic amino acids by clostridia. Arch Microbiol. 1976;107(3):283–288. doi: 10.1007/BF00425340.
169. Devlin AS, Marcobal A, Dodd D, Nayfach S, Plummer N, Meyer T, Pollard KS, Sonnenburg JL, Fischbach MA. Modulation of a circulating uremic solute via rational genetic manipulation of the gut microbiota. Cell Host & Microbe. 2016;20(6):709–715. doi: 10.1016/j.chom.2016.10.021.
170. Sokol H, Pigneur B, Watterlot L, Lakhdari O, Bermudez-Humaran LG, Gratadoux JJ, Blugeon S, Bridonneau C, Furet J-P, Corthier G, et al. Faecalibacterium prausnitzii is an anti-inflammatory commensal bacterium identified by gut microbiota analysis of Crohn disease patients. Proc Natl Acad Sci USA. 2008;105(43):16731–16736. doi: 10.1073/pnas.0804812105.
171. Aragozzini F, Ferrari A, Pacini N, Gualandris R. Indole-3-lactic acid as a tryptophan metabolite produced by bifidobacterium spp. Appl Environ Microbiol. 1979;38(3):544–546. doi: 10.1128/aem.38.3.544-546.1979.
172. Cervantes-Barragan L, Chai JN, Tianero MD, Di Luccia B, Ahern PP, Merriman J, Cortez VS, Caparon MG, Donia MS, Gilfillan S, et al. Lactobacillus reuteri induces gut intraepithelial CD4 + CD8αα + T cells. Science. 2017;357(6353):806–810. doi: 10.1126/science.aah5825.
173. Dodd D, Spitzer MH, Van Treuren W, Merrill BD, Hryckowian AJ, Higginbottom SK, Le A, Cowan TM, Nolan GP, Fischbach MA, et al. A gut bacterial pathway metabolizes aromatic amino acids into nine circulating metabolites. Nature. 2017;551(7682):648–652. doi: 10.1038/nature24661.
174. Honore AH, Aunsbjerg SD, Ebrahimi P, Thorsen M, Benfeldt C, Knochel S, Skov T. Metabolic footprinting for investigation of antifungal properties of Lactobacillus paracasei. Anal Bioanal Chem. 2016;408(1):83–96. doi: 10.1007/s00216-015-9103-6.
175. Russell WR, Duncan SH, Scobbie L, Duncan G, Cantlay L, Calder AG, Anderson SE, Flint HJ. Major phenylpropanoid-derived metabolites in the human gut can arise from microbial fermentation of protein. Mol Nutr Food Res. 2013;57(3):523–535. doi: 10.1002/mnfr.201200594.
176. Wilck N, Matus MG, Kearney SM, Olesen SW, Forslund K, Bartolomaeus H, Haase S, Mähler A, Balogh A, Markó L, et al. Salt-responsive gut commensal modulates TH17 axis and disease. Nature. 2017;551(7682):585–589. doi: 10.1038/nature24628.
177. Li D, Ni H, Jiao S, Lu Y, Zhou J, Sun B, Liang Y. Coexistence patterns of soil methanogens are closely tied to methane generation and community assembly in rice paddies. Microbiome. 2021;9(1):20. doi: 10.1186/s40168-020-00978-8.
178. Barbeyron T, Thomas F, Barbe V, Teeling H, Schenowitz C, Dossat C, Goesmann A, Leblanc C, Oliver Glöckner F, Czjzek M, et al. Habitat and taxon as driving forces of carbohydrate catabolism in marine heterotrophic bacteria: example of the model algae-associated bacterium zobellia galactanivorans dsij T. Environment Microbiol. 2016;18(12):4610–4627. doi: 10.1111/1462-2920.13584.

179. Valles-Colomer M, Falony G, Darzi Y, Tigchelaar EF, Wang J, Tito RY, Schiweck C, Kurilshikov A, Joossens M, Wijmenga C, et al. The neuroactive potential of the human gut microbiota in quality of life and depression. Nat Microbiol. 2019;4(4):623–632. doi: 10.1038/s41564-018-0337-x.
180. Zhu S, Wu Y, Zhu CY, Hong WC, Yu ZX, Chen ZK, Chen Z-L, Jiang D-G, Wang Y-G. The immediate mental health impacts of the COVID-19 pandemic among people with or without quarantine managements. Brain, Behavior, And Immunity. 2020;87:56–58. doi: 10.1016/j.bbi.2020.04.045.
181. Spanogiannopoulos P, Bess EN, Carmody RN, Turnbaugh PJ. The microbial pharmacists within us: a metagenomic view of xenobiotic metabolism. Nat Rev Microbiol. 2016;14(5):273–287. doi: 10.1038/nrmicro.2016.17.
182. Honeyfield DC, Carlson JR. Effect of indoleacetic acid and related indoles on Lactobacillus sp. Strain 11201 growth, indoleacetic acid catabolism, and 3-methylindole formation. Appl Environ Microbiol. 1990;56(5):1373–1377. doi: 10.1128/aem.56.5.1373-1377.1990.
183. Whitehead TR, Price NP, Drake HL, Cotta MA. Catabolic pathway for the production of skatole and indoleacetic acid by the acetogen clostridium drakei, clostridium scatologenes, and swine manure. Appl Environ Microbiol. 2008;74(6):1950–1953. doi: 10.1128/AEM.02458-07.
184. Wlodarska M, Luo C, Kolde R, d'Hennezel E, Annand JW, Heim CE, Krastel P, Schmitt EK, Omar AS, Creasey EA. Indoleacrylic acid produced by commensal peptostreptococcus species suppresses inflammation. Cell Host & Microbe. 2017;22(1):25–37.e6. doi: 10.1016/j.chom.2017.06.007.
185. Williams BB, Van Benschoten AH, Cimermancic P, Donia MS, Zimmermann M, Taketani M, Ishihara A, Kashyap P, Fraser J, Fischbach M. Discovery and characterization of gut microbiota decarboxylases that can produce the neurotransmitter tryptamine. Cell Host & Microbe. 2014;16(4):495–503. doi: 10.1016/j.chom.2014.09.001.
186. Sjogren K, Engdahl C, Henning P, Lerner UH, Tremaroli V, Lagerquist MK, Bäckhed F, Ohlsson C. The gut microbiota regulates bone mass in mice. J Of Bone And Mineral Res. 2012;27(6):1357–1367. doi: 10.1002/jbmr.1588.
187. Sanidad KZ, Rager SL, Carrow HC, Ananthanarayanan A, Callaghan R, Hart LR, Li T, Ravisankar P, Brown JA, Amir M, et al. Gut bacteria–derived serotonin promotes immune tolerance in early life. Sci Immunol. 2024;9(93):eadj4775. doi: 10.1126/sciimmunol.adj4775 .
188. Toh ML, Miossec P. The role of T cells in rheumatoid arthritis: new subsets and new targets. Curr Opin In Rheumatol. 2007;19(3):284–288. doi: 10.1097/BOR.0b013e32805e87e0.
189. Chabbi-Achengli Y, Coman T, Collet C, Callebert J, Corcelli M, Lin H, Rignault R, Dy M, de Vernejoul M-C, Côté F. Serotonin is involved in autoimmune arthritis through Th17 immunity and bone resorption. American J Of Pathol. 2016;186(4):927–937. doi: 10.1016/j.ajpath.2015.11.018.
190. Yang G, Wu G, Yao W, Guan L, Geng X, Liu J, Liu Z, Yang L, Huang Q, Zeng X, et al. 5-HT is associated with the dysfunction of regulating T cells in patients with allergic rhinitis. Clin Immunol. 2022;243:109101. doi: 10.1016/j.clim.2022.109101.
191. Gao J, Xu K, Liu H, Liu G, Bai M, Peng C, Li T, Yin Y. Impact of the gut microbiota on intestinal immunity mediated by tryptophan metabolism. Front Cell Infect Microbiol. 2018;8:13. doi: 10.3389/fcimb.2018.00013.
192. Lee JH, Wood TK, Lee J. Roles of indole as an interspecies and interkingdom signaling molecule. Trends Microbiol. 2015;23(11):707–718. doi: 10.1016/j.tim.2015.08.001.
193. Eme L, Gentekaki E, Curtis B, Archibald JM, Roger AJ. Lateral gene transfer in the adaptation of the anaerobic parasite blastocystis to the gut. Curr Biol. 2017;27(6):807–820. doi: 10.1016/j.cub.2017.02.003.
194. Yason JA, Liang YR, Png CW, Zhang Y, Tan KSW. Interactions between a pathogenic blastocystis subtype and gut microbiota: in vitro and in vivo studies. Microbiome. 2019;7(1):30. doi: 10.1186/s40168-019-0644-3.
195. Dong F, Perdew GH. The aryl hydrocarbon receptor as a mediator of host-microbiota interplay. Gut Microbes. 2020;12(1):1859812. doi: 10.1080/19490976.2020.1859812.
196. Tennoune N, Andriamihaja M, Blachier F. Production of indole and indole-related compounds by the intestinal microbiota and consequences for the host: the good, the bad, and the ugly. Microorganisms. 2022;10(5):930. doi: 10.3390/microorganisms10050930.
197. Lamas B, Richard ML, Leducq V, Pham HP, Michel ML, Da Costa G, Bridonneau C, Jegou S, Hoffmann TW, Natividad JM, et al. CARD9 impacts colitis by altering gut microbiota metabolism of tryptophan into aryl hydrocarbon receptor ligands. Nat Med. 2016;22(6):598–605. doi: 10.1038/nm.4102.
198. Michaudel C, Danne C, Agus A, Magniez A, Aucouturier A, Spatz M, Lefevre A, Kirchgesner J, Rolhion N, Wang Y, et al. Rewiring the altered tryptophan metabolism as a novel therapeutic strategy in inflammatory bowel diseases. Gut. 2023;72(7):1296–1307. doi: 10.1136/gutjnl-2022-327337.
199. Lucas LN, Barrett K, Kerby RL, Zhang Q, Cattaneo LE, Stevenson D, Rey FE, Amador-Noguez D. Dominant bacterial phyla from the human gut show widespread ability to transform and conjugate bile acids. mSystems. 2021;6(4):e0080521. doi: 10.1128/msystems.00805-21.
200. Wu CC, Weng WL, Lai WL, Tsai HP, Liu WH, Lee MH, Tsai Y-C. Effect of lactobacillus plantarum strain K21 on high-fat diet-fed obese mice. Evid Based Complement Alternat Med. 2015;2015:1–9. doi: 10.1155/2015/391767.

201. Ridlon JM, Ikegawa S, Alves JM, Zhou B, Kobayashi A, Iida T, Mitamura K, Tanabe G, Serrano M, De Guzman A, et al. Clostridium scindens: a human gut microbe with a high potential to convert glucocorticoids into androgens. J Of Lipid Res. 2013;54(9):2437–2449. doi: 10.1194/jlr.M038869.
202. Wahlstrom A, Brumbaugh A, Sjoland W, Olsson L, Wu H, Henricsson M, Lundqvist A, Makki K, Hazen SL, Bergström G. Production of deoxycholic acid by low-abundant microbial species is associated with impaired glucose metabolism. Nat Commun. 2024;15(1):4276. doi: 10.1038/s41467-024-48543-3.
203. Doden HL, Wolf PG, Gaskins HR, Anantharaman K, Alves JMP, Ridlon JM. Completion of the gut microbial epi-bile acid pathway. Gut Microbes. 2021;13(1):1–20. doi: 10.1080/19490976.2021.1907271.
204. Culpepper T, Rowe CC, Rusch CT, Burns AM, Federico AP, Girard SA, Tompkins TA, Nieves C, Dennis-Wall JC, Christman MC, et al. Three probiotic strains exert different effects on plasma bile acid profiles in healthy obese adults: randomised, double-blind placebo-controlled crossover study. BM. 2019;10(5):497–510. doi: 10.3920/BM2018.0151.
205. Sun L, Zhang Y, Cai J, Rimal B, Rocha ER, Coleman JP, Zhang C, Nichols RG, Luo Y, Kim B, et al. Bile salt hydrolase in non-enterotoxigenic bacteroides potentiates colorectal cancer. Nat Commun. 2023;14(1):755. doi: 10.1038/s41467-023-36089-9.
206. Xu F, Hu XJ, Singh W, Geng W, Tikhonova IG, Lin J. The complex structure of bile salt hydrolase from Lactobacillus salivarius reveals the structural basis of substrate specificity. Sci Rep. 2019;9(1):12438. doi: 10.1038/s41598-019-48850-6.
207. Prete R, Long SL, Gallardo AL, Gahan CG, Corsetti A, Joyce SA. Beneficial bile acid metabolism from Lactobacillus plantarum of food origin. Sci Rep. 2020;10(1):1165. doi: 10.1038/s41598-020-58069-5.
208. Wu L, Zhou J, Zhou A, Lei Y, Tang L, Hu S, Wang S, Xiao X, Chen Q, Tu D, et al. Lactobacillus acidophilus ameliorates cholestatic liver injury through inhibiting bile acid synthesis and promoting bile acid excretion. Gut Microbes. 2024;16(1):2390176. doi: 10.1080/19490976.2024.2390176.
209. Harris SC, Devendran S, Mendez-Garcia C, Mythen SM, Wright CL, Fields CJ, Hernandez AG, Cann I, Hylemon PB, Ridlon JM. Bile acid oxidation by eggerthella lenta strains C592 and DSM 2243(T). Gut Microbes. 2018;9(6):1–17. doi: 10.1080/19490976.2018.1458180.
210. Adhikari AA, Seegar TCM, Ficarro SB, McCurry MD, Ramachandran D, Yao L, Chaudhari SN, Ndousse-Fetter S, Banks AS, Marto JA, et al. Development of a covalent inhibitor of gut bacterial bile salt hydrolases. Nat Chem Biol. 2020;16(3):318–326. doi: 10.1038/s41589-020-0467-3.
211. De Juan A, Segura E. Modulation of immune responses by nutritional ligands of aryl hydrocarbon receptor. Front Immunol. 2021;12:645168. doi: 10.3389/fimmu.2021.645168.
212. Quintana FJ, Basso AS, Iglesias AH, Korn T, Farez MF, Bettelli E, Caccamo M, Oukka M, Weiner HL. Control of T(reg) and T(H)17 cell differentiation by the aryl hydrocarbon receptor. Nature. 2008;453(7191):65–71. doi: 10.1038/nature06880.
213. Gutierrez-Vazquez C, Quintana FJ. Regulation of the immune response by the aryl hydrocarbon receptor. Immunity. 2018;48(1):19–33. doi: 10.1016/j.immuni.2017.12.012.
214. Smirnova A, Wincent E, Vikstrom Bergander L, Alsberg T, Bergman J, Rannug A, Rannug U. Evidence for new light-independent pathways for generation of the endogenous aryl hydrocarbon receptor agonist FICZ. Chem Res Toxicol. 2016;29(1):75–86. doi: 10.1021/acs.chemrestox.5b00416.
215. Kenison JE, Jhaveri A, Li Z, Khadse N, Tjon E, Tezza S, Nowakowska D, Plasencia A, Stanton VP, Sherr DH, et al. Tolerogenic nanoparticles suppress central nervous system inflammation. Proc Natl Acad Sci USA. 2020;117(50):32017–32028. doi: 10.1073/pnas.2016451117.
216. Ye J, Qiu J, Bostick JW, Ueda A, Schjerven H, Li S, Jobin C, Chen ZME, Zhou L. The aryl hydrocarbon receptor preferentially marks and promotes gut regulatory T cells. Cell Reports. 2017;21(8):2277–2290. doi: 10.1016/j.celrep.2017.10.114.
217. Yoshimatsu Y, Sujino T, Miyamoto K, Harada Y, Tanemoto S, Ono K, Umeda S, Yoshida K, Teratani T, Suzuki T, et al. Aryl hydrocarbon receptor signals in epithelial cells govern the recruitment and location of Helios+ Tregs in the gut. Cell Reports. 2022;39(6):110773. doi: 10.1016/j.celrep.2022.110773.
218. Zhang Q, Zhu Y, Lv C, Fang Y, Liao M, Xia Y, Wei Z, Dai Y. AhR activation promotes treg cell generation by enhancing Lkb1-mediated fatty acid oxidation via the Skp2/K63-ubiquitination pathway. Immunol. 2023;169(4):412–430. doi: 10.1111/imm.13638.
219. Beischlag TV, Luis Morales J, Hollingshead BD, Perdew GH. The aryl hydrocarbon receptor complex and the control of gene expression. Crit Rev Eukaryot Gene Expr. 2008;18(3):207–250. doi: 10.1615/CritRevEukarGeneExpr.v18.i3.20.
220. Stockinger B, Di Meglio P, Gialitakis M, Duarte JH. The aryl hydrocarbon receptor: multitasking in the immune system. Annu Rev Immunol. 2014;32(1):403–432. doi: 10.1146/annurev-immunol-032713-120245.
221. Gandhi R, Kumar D, Burns EJ, Nadeau M, Dake B, Laroni A, Kozoriz D, Weiner HL, Quintana FJ. Activation of the aryl hydrocarbon receptor induces human type 1 regulatory T cell–like and Foxp3+ regulatory T cells. Nat Immunol. 2010;11(9):846–853. doi: 10.1038/ni.1915.
222. Apetoh L, Quintana FJ, Pot C, Joller N, Xiao S, Kumar D, Burns EJ, Sherr DH, Weiner HL, Kuchroo VK. The aryl hydrocarbon receptor interacts with c-maf to

promote the differentiation of type 1 regulatory T cells induced by IL-27. Nat Immunol. 2010;11(9):854–861. doi: 10.1038/ni.1912.
223. Nguyen NT, Kimura A, Nakahama T, Chinen I, Masuda K, Nohara K, Fujii-Kuriyama Y, Kishimoto T. Aryl hydrocarbon receptor negatively regulates dendritic cell immunogenicity via a kynurenine-dependent mechanism. Proc Natl Acad Sci USA. 2010;107(46):19961–19966. doi: 10.1073/pnas.1014465107.
224. Wang G, Fan Y, Zhang G, Cai S, Ma Y, Yang L, Wang Y, Yu H, Qiao S, Zeng X. Microbiota-derived indoles alleviate intestinal inflammation and modulate microbiome by microbial cross-feeding. Microbiome. 2024;12(1):59. doi: 10.1186/s40168-024-01750-y.
225. Atarashi K, Tanoue T, Oshima K, Suda W, Nagano Y, Nishikawa H, Fukuda S, Saito T, Narushima S, Hase K, et al. Treg induction by a rationally selected mixture of clostridia strains from the human microbiota. Nature. 2013;500(7461):232–236. doi: 10.1038/nature12331.
226. Stephen-Victor E, Kuziel GA, Martinez-Blanco M, Jugder BE, Benamar M, Wang Z, Chen Q, Lozano GL, Abdel-Gadir A, Cui Y, et al. RELMβ sets the threshold for microbiome-dependent oral tolerance. Nature. 2025;638(8051):760–768. doi: 10.1038/s41586-024-08440-7.
227. Nikolaus S, Schulte B, Al-Massad N, Thieme F, Schulte DM, Bethge J, Rehman A, Tran F, Aden K, Häsler R, et al. Increased tryptophan metabolism is associated with activity of inflammatory bowel diseases. Gastroenterology. 2017;153(6):1504–1516.e2. doi: 10.1053/j.gastro.2017.08.028.
228. Lim CK, Bilgin A, Lovejoy DB, Tan V, Bustamante S, Taylor BV, Bessede A, Brew BJ, Guillemin GJ. Kynurenine pathway metabolomics predicts and provides mechanistic insight into multiple sclerosis progression. Sci Rep. 2017;7(1):41473. doi: 10.1038/srep41473.
229. Schiering C, Wincent E, Metidji A, Iseppon A, Li Y, Potocnik AJ, Omenetti S, Henderson CJ, Wolf CR, Nebert DW, et al. Feedback control of AHR signalling regulates intestinal immunity. Nature. 2017;542(7640):242–245. doi: 10.1038/nature21080.
230. Schroecksnadel K, Winkler C, Duftner C, Wirleitner B, Schirmer M, Fuchs D. Tryptophan degradation increases with stage in patients with rheumatoid arthritis. Clin Rheumatol. 2006;25(3):334–337. doi: 10.1007/s10067-005-0056-6.
231. Hubbard TD, Murray IA, Perdew GH. Indole and tryptophan metabolism: endogenous and dietary routes to ah receptor activation. Drug Metab Dispos. 2015;43(10):1522–1535. doi: 10.1124/dmd.115.064246.
232. Shapiro H, Kolodziejczyk AA, Halstuch D, Elinav E. Bile acids in glucose metabolism in health and disease. J Exp Med. 2018;215(2):383–396. doi: 10.1084/jem.20171965.
233. Godlewska U, Bulanda E, Wypych TP. Bile acids in immunity: bidirectional mediators between the host and the microbiota. Front Immunol. 2022;13:949033. doi: 10.3389/fimmu.2022.949033.
234. Ridlon JM, Harris SC, Bhowmik S, Kang DJ, Hylemon PB. Consequences of bile salt biotransformations by intestinal bacteria. Gut Microbes. 2016;7(1):22–39. doi: 10.1080/19490976.2015.1127483.
235. Li J, Dawson PA. Animal models to study bile acid metabolism. Biochim Biophys Acta Mol Basis Dis. 2019;1865(5):895–911. doi: 10.1016/j.bbadis.2018.05.011.
236. Marion S, Desharnais L, Studer N, Dong Y, Notter MD, Poudel S, Menin L, Janowczyk A, Hettich RL, Hapfelmeier S, et al. Biogeography of microbial bile acid transformations along the murine gut. J Of Lipid Res. 2020;61(11):1450–1463. doi: 10.1194/jlr.RA120001021.
237. Collins SL, Stine JG, Bisanz JE, Okafor CD, Patterson AD. Bile acids and the gut microbiota: metabolic interactions and impacts on disease. Nat Rev Microbiol. 2023;21(4):236–247. doi: 10.1038/s41579-022-00805-x.
238. Gustafsson BE, Midtvedt T, Norman A. Metabolism of cholic acid in germfree animals after the establishment in the intestinal tract of deconjugating and 7 alpha-dehydroxylating bacteria. Acta Pathologica Microbiologica Scandinavica. 1968;72(3):433–443. doi: 10.1111/j.1699-0463.1968.tb00457.x.
239. Narushima S, Itoha K, Miyamoto Y, Park SH, Nagata K, Kuruma K, Uchida K. Deoxycholic acid formation in gnotobiotic mice associated with human intestinal bacteria. Lipids. 2006;41(9):835–843. doi: 10.1007/s11745-006-5038-1.
240. van Best N, Rolle-Kampczyk U, Schaap FG, Basic M, Olde Damink SWM, Bleich A, Savelkoul PHM, von Bergen M, Penders J, Hornef MW. Bile acids drive the newborn's gut microbiota maturation. Nat Commun. 2020;11(1):3692. doi: 10.1038/s41467-020-17183-8.
241. Li Y, Tang R, Leung PSC, Gershwin ME, Ma X. Bile acids and intestinal microbiota in autoimmune cholestatic liver diseases. Autoimmun Rev. 2017;16(9):885–896. doi: 10.1016/j.autrev.2017.07.002.
242. Cabrera-Rubio R, Patterson AM, Cotter PD, Beraza N. Cholestasis induced by bile duct ligation promotes changes in the intestinal microbiome in mice. Sci Rep. 2019;9(1):12324. doi: 10.1038/s41598-019-48784-z.
243. Makishima M, Okamoto AY, Repa JJ, Tu H, Learned RM, Luk A, Hull MV, Lustig KD, Mangelsdorf DJ, Shan B. Identification of a nuclear receptor for bile acids. Science. 1999;284(5418):1362–1365. doi: 10.1126/science.284.5418.1362.
244. Parks DJ, Blanchard SG, Bledsoe RK, Chandra G, Consler TG, Kliewer SA, Stimmel JB, Willson TM, Zavacki AM, Moore DD, et al. Bile acids: natural ligands for an orphan nuclear receptor. Science. 1999;284(5418):1365–1368. doi: 10.1126/science.284.5418.1365.
245. Chiang JY. Bile acids: regulation of synthesis. J Lipid Res. 2009;50(10):1955–1966. doi: 10.1194/jlr.R900010-JLR200.

246. Maruyama T, Miyamoto Y, Nakamura T, Tamai Y, Okada H, Sugiyama E, Nakamura T, Itadani H, Tanaka K. Identification of membrane-type receptor for bile acids (M-BAR). Biochem And Biophysical Res Communicat. 2002;298(5):714–719. doi: 10.1016/S0006-291X(02)02550-0.
247. Makishima M, Lu TT, Xie W, Whitfield GK, Domoto H, Evans RM, Haussler MR, Mangelsdorf DJ. Vitamin D receptor as an intestinal bile acid sensor. Science. 2002;296(5571):1313–1316. doi: 10.1126/science.1070477.
248. Thibaut MM, Bindels LB. Crosstalk between bile acid-activated receptors and microbiome in entero-hepatic inflammation. Trends Mol Med. 2022;28(3):223–236. doi: 10.1016/j.molmed.2021.12.006.
249. Wagner M, Halilbasic E, Marschall HU, Zollner G, Fickert P, Langner C, Zatloukal K, Denk H, Trauner M. CAR and PXR agonists stimulate hepatic bile acid and bilirubin detoxification and elimination pathways in mice. Hepatology. 2005;42(2):420–430. doi: 10.1002/hep.20784.
250. Raufman JP, Cheng K, Zimniak P. Activation of muscarinic receptor signaling by bile acids: physiological and medical implications. Dig Dis Sci. 2003;48(8):1431–1444. doi: 10.1023/A:1024733500950.
251. Nagahashi M, Takabe K, Liu R, Peng K, Wang X, Wang Y, Hait NC, Wang X, Allegood JC, Yamada A, et al. Conjugated bile acid-activated S1P receptor 2 is a key regulator of sphingosine kinase 2 and hepatic gene expression. Hepatology. 2015;61(4):1216–1226. doi: 10.1002/hep.27592.
252. Li W, Hang S, Fang Y, Bae S, Zhang Y, Zhang M, Wang G, McCurry MD, Bae M, Paik D, et al. A bacterial bile acid metabolite modulates T(reg) activity through the nuclear hormone receptor NR4A1. Cell Host & Microbe. 2021;29(9):1366–1377.e9. doi: 10.1016/j.chom.2021.07.013.
253. Paik D, Yao L, Zhang Y, Bae S, D'Agostino GD, Zhang M, Kim E, Franzosa EA, Avila-Pacheco J, Bisanz JE, et al. Human gut bacteria produce τη17-modulating bile acid metabolites. Nature. 2022;603(7903):907–912. doi: 10.1038/s41586-022-04480-z.
254. Campbell C, McKenney PT, Konstantinovsky D, Isaeva OI, Schizas M, Verter J, Mai C, Jin W-B, Guo C-J, Violante S, et al. Bacterial metabolism of bile acids promotes generation of peripheral regulatory T cells. Nature. 2020;581(7809):475–479. doi: 10.1038/s41586-020-2193-0.
255. Song X, Sun X, Oh SF, Wu M, Zhang Y, Zheng W, Geva-Zatorsky N, Jupp R, Mathis D, Benoist C, et al. Microbial bile acid metabolites modulate gut RORγ+ regulatory T cell homeostasis. Nature. 2020;577(7790):410–415. doi: 10.1038/s41586-019-1865-0.
256. Duboc H, Rajca S, Rainteau D, Benarous D, Maubert MA, Quervain E, Thomas G, Barbu V, Humbert L, Despras G, et al. Connecting dysbiosis, bile-acid dysmetabolism and gut inflammation in inflammatory bowel diseases. Gut. 2013;62(4):531–539. doi: 10.1136/gutjnl-2012-302578.
257. Thomas JP, Modos D, Rushbrook SM, Powell N, Korcsmaros T. The emerging role of bile acids in the pathogenesis of inflammatory bowel disease. Front Immunol. 2022;13:829525. doi: 10.3389/fimmu.2022.829525.
258. Tiratterra E, Franco P, Porru E, Katsanos KH, Christodoulou DK, Roda G. Role of bile acids in inflammatory bowel disease. Ann Gastroenterol. 2018;31(3):266–272. doi: 10.20524/aog.2018.0239.
259. Fickert P, Wagner M. Biliary bile acids in hepatobiliary injury - what is the link? J Of Hepatol. 2017;67(3):619–631. doi: 10.1016/j.jhep.2017.04.026.
260. Sayin SI, Wahlstrom A, Felin J, Jantti S, Marschall HU, Bamberg K, Angelin B, Hyötyläinen T, Orešič M, Bäckhed F. Gut microbiota regulates bile acid metabolism by reducing the levels of tauro-beta-muricholic acid, a naturally occurring FXR antagonist. Cell Metabol. 2013;17(2):225–235. doi: 10.1016/j.cmet.2013.01.003.
261. Winston JA, Theriot CM. Diversification of host bile acids by members of the gut microbiota. Gut Microbes. 2020;11(2):158–171. doi: 10.1080/19490976.2019.1674124.
262. Fiorucci S, Biagioli M, Zampella A, Distrutti E. Bile acids activated receptors regulate innate immunity. Front Immunol. 2018;9:1853. doi: 10.3389/fimmu.2018.01853.
263. Ridlon JM, Kang DJ, Hylemon PB, Bajaj JS. Bile acids and the gut microbiome. Curr Opin In Gastroenterol. 2014;30(3):332–338. doi: 10.1097/MOG.0000000000000057.
264. Cao H, Xu M, Dong W, Deng B, Wang S, Zhang Y, Wang S, Luo S, Wang W, Qi Y, et al. Secondary bile acid-induced dysbiosis promotes intestinal carcinogenesis. Int J Cancer. 2017;140(11):2545–2556. doi: 10.1002/ijc.30643.
265. Yoshimoto S, Loo TM, Atarashi K, Kanda H, Sato S, Oyadomari S, Iwakura Y, Oshima K, Morita H, Hattori M, et al. Obesity-induced gut microbial metabolite promotes liver cancer through senescence secretome. Nature. 2013;499(7456):97–101. doi: 10.1038/nature12347.
266. Maharshak N, Packey CD, Ellermann M, Manick S, Siddle JP, Huh EY, Plevy S, Sartor RB, Carroll IM. Altered enteric microbiota ecology in interleukin 10-deficient mice during development and progression of intestinal inflammation. Gut Microbes. 2013;4(4):316–324. doi: 10.4161/gmic.25486.
267. Koshida K, Ito M, Yakabe K, Takahashi Y, Tai Y, Akasako R, Kimizuka T, Takano S, Sakamoto N, Haniuda K, et al. Dysfunction of Foxp3+ regulatory T cells induces dysbiosis of gut microbiota via aberrant binding of immunoglobulins to microbes in the intestinal lumen. Int J Mol Sci. 2023;24(10):8549. doi: 10.3390/ijms24108549.

268. Lochner M, Berard M, Sawa S, Hauer S, Gaboriau-Routhiau V, Fernandez TD, Snel J, Bousso P, Cerf-Bensussan N, Eberl G. Restricted microbiota and absence of cognate TCR antigen leads to an unbalanced generation of Th17 cells. The J Of Immunol. 2011;186 (3):1531–1537. doi: 10.4049/jimmunol.1001723.
269. d'Hennezel E, Bin Dhuban K, Torgerson T, Piccirillo CA. The immunogenetics of immune dysregulation, polyendocrinopathy, enteropathy, X linked (IPEX) syndrome. J Med Genet. 2012;49(5):291–302. doi: 10.1136/jmedgenet-2012-100759.
270. Vujkovic-Cvijin I, Dunham RM, Iwai S, Maher MC, Albright RG, Broadhurst MJ, Hernandez RD, Lederman MM, Huang Y, Somsouk M, et al. Dysbiosis of the gut microbiota is associated with HIV disease progression and tryptophan catabolism. Sci Transl Med. 2013;5(193):193ra91. doi: 10.1126/scitranslmed.3006438.
271. Tourret J, Willing BP, Dion S, MacPherson J, Denamur E, Finlay BB. Immunosuppressive treatment alters secretion of ileal antimicrobial peptides and gut microbiota, and favors subsequent colonization by uropathogenic *Escherichia coli*. Transplantation. 2017;101(1):74–82. doi: 10.1097/TP.0000000000001492.
272. Kuhn KA, Stappenbeck TS. Peripheral education of the immune system by the colonic microbiota. Semin Immunol. 2013;25(5):364–369. doi: 10.1016/j.smim.2013.10.002.
273. Pickard JM, Zeng MY, Caruso R, Nunez G. Gut microbiota: role in pathogen colonization, immune responses, and inflammatory disease. Immunol Rev. 2017;279(1):70–89. doi: 10.1111/imr.12567.
274. Sartor RB. Therapeutic manipulation of the enteric microflora in inflammatory bowel diseases: antibiotics, probiotics, and prebiotics. Gastroenterology. 2004;126 (6):1620–1633. doi: 10.1053/j.gastro.2004.03.024.
275. Huang H, Fang M, Jostins L, Umicevic Mirkov M, Boucher G, Anderson CA, Andersen V, Cleynen I, Cortes A, Crins F, et al. Fine-mapping inflammatory bowel disease loci to single-variant resolution. Nature. 2017;547(7662):173–178. doi: 10.1038/nature22969.
276. Jostins L, Ripke S, Weersma RK, Duerr RH, McGovern DP, Hui KY, Lee JC, Philip Schumm L, Sharma Y, Anderson CA, et al. Host-microbe interactions have shaped the genetic architecture of inflammatory bowel disease. Nature. 2012;491(7422):119–124. doi: 10.1038/nature11582.
277. Franzosa EA, Sirota-Madi A, Avila-Pacheco J, Fornelos N, Haiser HJ, Reinker S, Vatanen T, Hall AB, Mallick H, McIver LJ, et al. Gut microbiome structure and metabolic activity in inflammatory bowel disease. Nat Microbiol. 2019;4(2):293–305. doi: 10.1038/s41564-018-0306-4.
278. Lee M, Chang EB. Inflammatory bowel diseases (IBD) and the microbiome—searching the crime scene for clues. Gastroenterology. 2021;160(2):524–537. doi: 10.1053/j.gastro.2020.09.056.
279. Kostic AD, Xavier RJ, Gevers D. The microbiome in inflammatory bowel disease: current status and the future ahead. Gastroenterology. 2014;146(6):1489–1499. doi: 10.1053/j.gastro.2014.02.009.
280. Pasolli E, Asnicar F, Manara S, Zolfo M, Karcher N, Armanini F, Beghini F, Manghi P, Tett A, Ghensi P, et al. Extensive unexplored human microbiome diversity revealed by over 150,000 genomes from metagenomes spanning age, geography, and lifestyle. Cell. 2019;176 (3):649–662.e20. doi: 10.1016/j.cell.2019.01.001.
281. Kruis W, Kalek HD, Stellaard F, Paumgartner G. Altered fecal bile acid pattern in patients with inflammatory bowel disease. Digestion. 1986;35(4):189–198. doi: 10.1159/000199367.
282. Ott SJ, Kuhbacher T, Musfeldt M, Rosenstiel P, Hellmig S, Rehman A, Drews O, Weichert W, Timmis KN, Schreiber S. Fungi and inflammatory bowel diseases: alterations of composition and diversity. Scandinavian J Of Gastroenterol. 2008;43(7):831–841. doi: 10.1080/00365520801935434.
283. Richard ML, Lamas B, Liguori G, Hoffmann TW, Sokol H. Gut fungal microbiota: the Yin and Yang of inflammatory bowel disease. Inflamm Bowel Dis. 2015;21 (3):656–665. doi: 10.1097/MIB.0000000000000261.
284. Britton GJ, Contijoch EJ, Mogno I, Vennaro OH, Llewellyn SR, Ng R, Li Z, Mortha A, Merad M, Das A, et al. Microbiotas from humans with inflammatory bowel disease alter the balance of gut Th17 and RORgammat(+) regulatory T cells and exacerbate colitis in mice. Immunity. 2019;50(1):212–224.e4. doi: 10.1016/j.immuni.2018.12.015.
285. Maul J, Loddenkemper C, Mundt P, Berg E, Giese T, Stallmach A, Zeitz M, Duchmann R. Peripheral and intestinal regulatory CD4+CD25high T cells in inflammatory bowel disease. Gastroenterology. 2005;128 (7):1868–1878. doi: 10.1053/j.gastro.2005.03.043.
286. Saruta M, Yu QT, Fleshner PR, Mantel PY, Schmidt-Weber CB, Banham AH, Papadakis KA. Characterization of FOXP3+CD4+ regulatory T cells in Crohn's disease. Clin Immunoly. 2007;125(3):281–290. doi: 10.1016/j.clim.2007.08.003.
287. Reikvam DH, Perminow G, Lyckander LG, Gran JM, Brandtzaeg P, Vatn M, Carlsen HS. Increase of regulatory T cells in ileal mucosa of untreated pediatric Crohn's disease patients. Scandinavian J Of Gastroenterol. 2011;46(5):550–560. doi: 10.3109/00365521.2011.551887.
288. Goldberg R, Scotta C, Cooper D, Nissim-Eliraz E, Nir E, Tasker S, Irving PM, Sanderson J, Lavender P, Ibrahim F, et al. Correction of defective T-Regulatory cells from patients with Crohn's disease by ex vivo ligation of retinoic acid receptor-α. Gastroenterology. 2019;156(6):1775–1787. doi: 10.1053/j.gastro.2019.01.025.
289. Lord JD, Shows DM, Chen J, Thirlby RC, Unutmaz D. Human blood and mucosal regulatory T cells express activation markers and inhibitory receptors in

inflammatory bowel disease. PLOS ONE. 2015;10(8): e0136485. doi: 10.1371/journal.pone.0136485.
290. Fantini MC, Rizzo A, Fina D, Caruso R, Sarra M, Stolfi C, Becker C, MacDonald TT, Pallone F, Neurath MF, et al. Smad7 controls resistance of colitogenic T cells to regulatory T cell-mediated suppression. Gastroenterology. 2009;136(4):1308–1316.e3. doi: 10.1053/j.gastro.2008.12.053.
291. Mitsialis V, Wall S, Liu P, Ordovas-Montanes J, Parmet T, Vukovic M, Spencer D, Field M, McCourt C, Toothaker J, et al. Single-cell analyses of colon and blood reveal distinct immune cell signatures of ulcerative colitis and Crohn's disease. Gastroenterology. 2020;159(2):591–608.e10. doi: 10.1053/j.gastro.2020.04.074.
292. Smillie CS, Biton M, Ordovas-Montanes J, Sullivan KM, Burgin G, Graham DB, Herbst RH, Rogel N, Slyper M, Waldman J, et al. Intra- and inter-cellular rewiring of the human colon during ulcerative colitis. Cell. 2019;178(3):714–730.e22. doi: 10.1016/j.cell.2019.06.029.
293. Allegretti JR, Mitsialis V, Canavan JB, Low-Dose ILUCSG, Snapper SB, Barends J, Carrellas M, Freer K, Gringauz J, Green J, et al. Low-dose interleukin 2 for the treatment of moderate to severe ulcerative colitis. Gastroenterology. 2023;165(2):492–495.e2. doi: 10.1053/j.gastro.2023.03.230.
294. Allegretti JR, Kelly CR, Grinspan A, Mullish BH, Hurtado J, Carrellas M, Marcus J, Marchesi JR, McDonald JAK, Gerardin Y, et al. Inflammatory bowel disease outcomes following fecal microbiota transplantation for recurrent C. difficile infection. Inflammat Bowel Dis. 2021;27(9):1371–1378. doi: 10.1093/ibd/izaa283.
295. Haifer C, Paramsothy S, Kaakoush NO, Saikal A, Ghaly S, Yang T, Luu LDW, Borody TJ, Leong RW. Lyophilised oral faecal microbiota transplantation for ulcerative colitis (LOTUS): a randomised, double-blind, placebo-controlled trial. Lancet Gastroenterol Hepatol. 2022;7(2):141–151. doi: 10.1016/S2468-1253(21)00400-3.
296. Pai N, Popov J, Hill L, Hartung E, Grzywacz K, Moayyedi P, Surette M, Lee C, Godin D, Szamosi JC, et al. Results of the first pilot randomized controlled trial of fecal microbiota transplant in pediatric ulcerative colitis: lessons, limitations, and future prospects. Gastroenterology. 2021;161(2):388–393.e3. doi: 10.1053/j.gastro.2021.04.067.
297. Leonard MM, Serena G, Sturgeon C, Fasano A. Genetics and celiac disease: the importance of screening. Expert Rev Gastroenterol Hepatol. 2015;9(2):209–215. doi: 10.1586/17474124.2014.945915.
298. Wacklin P, Kaukinen K, Tuovinen E, Collin P, Lindfors K, Partanen J, Mäki M, Mättö J. The duodenal microbiota composition of adult celiac disease patients is associated with the clinical manifestation of the disease. Inflammat Bowel Dis. 2013;19(5):934–941. doi: 10.1097/MIB.0b013e31828029a9.
299. Fernandez S, Molina IJ, Romero P, Gonzalez R, Pena J, Sanchez F, Reynoso FR, Pérez-Navero JL, Estevez O, Ortega C, et al. Characterization of gliadin-specific Th17 cells from the mucosa of celiac disease patients. American J Of Gastroenterol. 2011;106(3):528–538. doi: 10.1038/ajg.2010.465.
300. Nilsen EM, Jahnsen FL, Lundin KE, Johansen FE, Fausa O, Sollid LM, Jahnsen J, Scott H, Brandtzaeg P. Gluten induces an intestinal cytokine response strongly dominated by interferon gamma in patients with celiac disease. Gastroenterology. 1998;115(3):551–563. doi: 10.1016/S0016-5085(98)70134-9.
301. Borrelli M, Salvati VM, Maglio M, Zanzi D, Ferrara K, Santagata S, Ponticelli D, Aitoro R, Mazzarella G, Lania G, et al. Immunoregulatory pathways are active in the small intestinal mucosa of patients with potential celiac disease. American J Of Gastroenterol. 2013;108(11):1775–1784. doi: 10.1038/ajg.2013.303.
302. Forsberg G, Hernell O, Melgar S, Israelsson A, Hammarstrom S, Hammarstrom ML. Paradoxical coexpression of proinflammatory and down-regulatory cytokines in intestinal T cells in childhood celiac disease. Gastroenterology. 2002;123(3):667–678. doi: 10.1053/gast.2002.35355.
303. Salvati VM, Mazzarella G, Gianfrani C, Levings MK, Stefanile R, De Giulio B. Recombinant human interleukin 10 suppresses gliadin dependent T cell activation in ex vivo cultured coeliac intestinal mucosa. Gut. 2005;54(1):46–53. doi: 10.1136/gut.2003.023150.
304. Hmida NB, Ben Ahmed M, Moussa A, Rejeb MB, Said Y, Kourda N, Meresse B, Abdeladhim M, Louzir H, Cerf-Bensussan N. Impaired control of effector T cells by regulatory T cells: a clue to loss of oral tolerance and autoimmunity in celiac disease? American J Of Gastroenterol. 2012;107(4):604–611. doi: 10.1038/ajg.2011.397.
305. Tiittanen M, Westerholm-Ormio M, Verkasalo M, Savilahti E, Vaarala O. Infiltration of forkhead box P3-expressing cells in small intestinal mucosa in coeliac disease but not in type 1 diabetes. Clin Exp Immunol. 2008;152(3):498–507. doi: 10.1111/j.1365-2249.2008.03662.x.
306. Vorobjova T, Uibo O, Heilman K, Rago T, Honkanen J, Vaarala O, Tillmann V, Ojakivi I, Uibo R. Increased FOXP3 expression in small-bowel mucosa of children with coeliac disease and type I diabetes mellitus. Scandinavian J Of Gastroenterol. 2009;44(4):422–430. doi: 10.1080/00365520802624177.
307. Granzotto M, Dal Bo S, Quaglia S, Tommasini A, Piscianz E, Valencic E, Ferrara F, Martelossi S, Ventura A, Not T. Regulatory T-cell function is impaired in celiac disease. Dig Dis Sci. 2009;54(7):1513–1519. doi: 10.1007/s10620-008-0501-x.
308. Cook L, Munier CML, Seddiki N, van Bockel D, Ontiveros N, Hardy MY, Gillies JK, Levings MK, Reid HH, Petersen J, et al. Circulating gluten-specific FOXP3 (+)CD39(+) regulatory T cells have impaired

suppressive function in patients with celiac disease. J Of Allergy And Clin Immunol. 2017;140(6):1592–1603.e8. doi: 10.1016/j.jaci.2017.02.015.
309. Ben Ahmed M, Belhadj Hmida N, Moes N, Buyse S, Abdeladhim M, Louzir H, Cerf-Bensussan N. IL-15 renders conventional lymphocytes resistant to suppressive functions of regulatory T cells through activation of the phosphatidylinositol 3-kinase pathway. J Of Immunol. 2009;182(11):6763–6770. doi: 10.4049/jimmunol.0801792.
310. Benahmed M, Meresse B, Arnulf B, Barbe U, Mention JJ, Verkarre V, Allez M, Cellier C, Hermine O, Cerf–Bensussan N. Inhibition of TGF-β signaling by IL-15: a new role for IL-15 in the loss of immune homeostasis in celiac disease. Gastroenterology. 2007;132(3):994–1008. doi: 10.1053/j.gastro.2006.12.025.
311. Serena G, Yan S, Camhi S, Patel S, Lima RS, Sapone A, Leonard MM, Mukherjee R, Nath BJ, Lammers KM, et al. Proinflammatory cytokine interferon-gamma and microbiome-derived metabolites dictate epigenetic switch between forkhead box protein 3 isoforms in coeliac disease. Clin And Experimental Immunol. 2017;187(3):490–506. doi: 10.1111/cei.12911.
312. Du J, Huang C, Zhou B, Ziegler SF. Isoform-specific inhibition of RORα-mediated transcriptional activation by human FOXP3. The J Of Immunol. 2008;180(7):4785–4792. doi: 10.4049/jimmunol.180.7.4785.
313. De Palma G, Capilla A, Nadal I, Nova E, Pozo T, Varea V, Polanco I, Castillejo G, López A, Garrote JA, et al. Interplay between human leukocyte antigen genes and the microbial colonization process of the newborn intestine. Curr Issues Mol Biol. 2010;12(1):1–10.
314. Olivares M, Neef A, Castillejo G, Palma GD, Varea V, Capilla A, Palau F, Nova E, Marcos A, Polanco I, et al. The HLA-DQ2 genotype selects for early intestinal microbiota composition in infants at high risk of developing coeliac disease. Gut. 2015;64(3):406–417. doi: 10.1136/gutjnl-2014-306931.
315. Sellitto M, Bai G, Serena G, Fricke WF, Sturgeon C, Gajer P, White JR, Koenig SSK, Sakamoto J, Boothe D, et al. Proof of concept of microbiome-metabolome analysis and delayed gluten exposure on celiac disease autoimmunity in genetically at-risk infants. PLOS ONE. 2012;7(3):e33387. doi: 10.1371/journal.pone.0033387.
316. Wong SH, Zhao L, Zhang X, Nakatsu G, Han J, Xu W, Xiao X, Kwong TNY, Tsoi H, Wu WKK, et al. Gavage of fecal samples from patients with colorectal cancer promotes intestinal carcinogenesis in germ-free and conventional mice. Gastroenterology. 2017;153(6):1621–1633.e6. doi: 10.1053/j.gastro.2017.08.022.
317. Dai Z, Coker OO, Nakatsu G, Wu WKK, Zhao L, Chen Z, Chan FKL, Kristiansen K, Sung JJY, Wong SH, et al. Multi-cohort analysis of colorectal cancer metagenome identified altered bacteria across populations and universal bacterial markers. Microbiome. 2018;6(1):70. doi: 10.1186/s40168-018-0451-2.
318. Lee KA, Thomas AM, Bolte LA, Bjork JR, de Ruijter LK, Armanini F, Asnicar F, Blanco-Miguez A, Board R, Calbet-Llopart N, et al. Cross-cohort gut microbiome associations with immune checkpoint inhibitor response in advanced melanoma. Nat Med. 2022;28(3):535–544. doi: 10.1038/s41591-022-01695-5.
319. McCulloch JA, Davar D, Rodrigues RR, Badger JH, Fang JR, Cole AM, Balaji AK, Vetizou M, Prescott SM, Fernandes MR, et al. Intestinal microbiota signatures of clinical response and immune-related adverse events in melanoma patients treated with anti-PD-1. Nat Med. 2022;28(3):545–556. doi: 10.1038/s41591-022-01698-2.
320. Gunjur A, Shao Y, Rozday T, Klein O, Mu A, Haak BW, Markman B, Kee D, Carlino MS, Underhill C, et al. A gut microbial signature for combination immune checkpoint blockade across cancer types. Nat Med. 2024;30(3):797–809. doi: 10.1038/s41591-024-02823-z.
321. Peng Z, Cheng S, Kou Y, Wang Z, Jin R, Hu H, Zhang X, Gong J-F, Li J, Lu M, et al. The gut microbiome is associated with clinical response to Anti-PD-1/PD-L1 immunotherapy in gastrointestinal cancer. Cancer Immunol Res. 2020;8(10):1251–1261. doi: 10.1158/2326-6066.CIR-19-1014.
322. Thomas AM, Manghi P, Asnicar F, Pasolli E, Armanini F, Zolfo M, Beghini F, Manara S, Karcher N, Pozzi C, et al. Metagenomic analysis of colorectal cancer datasets identifies cross-cohort microbial diagnostic signatures and a link with choline degradation. Nat Med. 2019;25(4):667–678. doi: 10.1038/s41591-019-0405-7.
323. Wirbel J, Pyl PT, Kartal E, Zych K, Kashani A, Milanese A, Fleck JS, Voigt AY, Palleja A, Ponnudurai R, et al. Meta-analysis of fecal metagenomes reveals global microbial signatures that are specific for colorectal cancer. Nat Med. 2019;25(4):679–689. doi: 10.1038/s41591-019-0406-6.
324. Yachida S, Mizutani S, Shiroma H, Shiba S, Nakajima T, Sakamoto T, Watanabe H, Masuda K, Nishimoto Y, Kubo M, et al. Metagenomic and metabolomic analyses reveal distinct stage-specific phenotypes of the gut microbiota in colorectal cancer. Nat Med. 2019;25(6):968–976. doi: 10.1038/s41591-019-0458-7.
325. Bernstein H, Bernstein C, Payne CM, Dvorak K. Bile acids as endogenous etiologic agents in gastrointestinal cancer. World J Gastroenterol. 2009;15(27):3329–3340. doi: 10.3748/wjg.15.3329.
326. Suzuki K, Bruce WR. Increase by deoxycholic acid of the colonic nuclear damage induced by known carcinogens in C57BL/6J mice. J Natl Cancer Inst. 1986;76:1129–1132.
327. Balkwill F, Mantovani A. Inflammation and cancer: back to Virchow? Lancet. 2001;357(9255):539–545. doi: 10.1016/S0140-6736(00)04046-0.
328. Ladoire S, Martin F, Ghiringhelli F. Prognostic role of FOXP3+ regulatory T cells infiltrating human carcinomas: the paradox of colorectal cancer. Cancer Immunol, Immunother: CII. 2011;60(7):909–918. doi: 10.1007/s00262-011-1046-y.

329. Pastille E, Bardini K, Fleissner D, Adamczyk A, Frede A, Wadwa M, von Smolinski D, Kasper S, Sparwasser T, Gruber AD, et al. Transient ablation of regulatory T cells improves antitumor immunity in colitis-associated colon cancer. Cancer Res. 2014;74(16):4258–4269. doi: 10.1158/0008-5472.CAN-13-3065.
330. Sharma A, Sharma G, Gao Z, Li K, Li M, Wu M, Kim CJ, Chen Y, Gautam A, Choi HB, et al. Glut3 promotes cellular O-GlcNAcylation as a distinctive tumor-supportive feature in treg cells. Cell Mol Immunol. 2024;21(12):1474–1490. doi: 10.1038/s41423-024-01229-8.
331. Cavalleri T, Bianchi P, Basso G, Celesti G, Grizzi F, Bossi P, Greco L, Pitrone C, Valtorta E, Mauri G, et al. Combined low densities of FoxP3+ and CD3+ tumor-infiltrating lymphocytes identify stage II colorectal cancer at high risk of progression. Cancer Immunol Res. 2019;7(5):751–758. doi: 10.1158/2326-6066.CIR-18-0661.
332. Salama P, Phillips M, Grieu F, Morris M, Zeps N, Joseph D, Platell C, Iacopetta B. Tumor-infiltrating FOXP3+ T regulatory cells show strong prognostic significance in colorectal cancer. JCO. 2009;27(2):186–192. doi: 10.1200/JCO.2008.18.7229.
333. Saito T, Nishikawa H, Wada H, Nagano Y, Sugiyama D, Atarashi K, Maeda Y, Hamaguchi M, Ohkura N, Sato E, et al. Two FOXP3(+)CD4(+) T cell subpopulations distinctly control the prognosis of colorectal cancers. Nat Med. 2016;22(6):679–684. doi: 10.1038/nm.4086.
334. Frei AL, McGuigan A, Sinha R, Jabbar F, Gneo L, Tomasevic T, Harkin A, Iveson T, Saunders MP, Oien KA, et al. Multiplex analysis of intratumoural immune infiltrate and prognosis in patients with stage II-III colorectal cancer from the SCOT and QUASAR 2 trials: a retrospective analysis. The Lancet Oncol. 2024;25(2):198–211. doi: 10.1016/S1470-2045(23)00560-0.
335. Zheng D, Liwinski T, Elinav E. Interaction between microbiota and immunity in health and disease. Cell Res. 2020;30(6):492–506. doi: 10.1038/s41422-020-0332-7.
336. Litvak Y, Byndloss MX, Tsolis RM, Baumler AJ. Dysbiotic proteobacteria expansion: a microbial signature of epithelial dysfunction. Curr Opin Microbiol. 2017;39:1–6. doi: 10.1016/j.mib.2017.07.003.
337. Kalliomaki M, Salminen S, Poussa T, Arvilommi H, Isolauri E. Probiotics and prevention of atopic disease: 4-year follow-up of a randomised placebo-controlled trial. Lancet. 2003;361(9372):1869–1871. doi: 10.1016/S0140-6736(03)13490-3.
338. Lewis JD, Sandler RS, Brotherton C, Brensinger C, Li H, Kappelman MD, Daniel SG, Bittinger K, Albenberg L, Valentine JF, et al. A randomized trial comparing the specific carbohydrate diet to a Mediterranean diet in adults with Crohn's disease. Gastroenterology. 2021;161(3):837–852.e9. doi: 10.1053/j.gastro.2021.05.047.
339. Walter J, Armet AM, Finlay BB, Shanahan F. Establishing or exaggerating causality for the gut microbiome: lessons from human microbiota-associated rodents. Cell. 2020;180(2):221–232. doi: 10.1016/j.cell.2019.12.025.
340. Klatzmann D, Abbas AK. The promise of low-dose interleukin-2 therapy for autoimmune and inflammatory diseases. Nat Rev Immunol. 2015;15(5):283–294. doi: 10.1038/nri3823.
341. Desreumaux P, Foussat A, Allez M, Beaugerie L, Hebuterne X, Bouhnik Y. Safety and efficacy of antigen-specific regulatory T-cell therapy for patients with refractory Crohn's disease. Gastroenterology. 2012;143(5):1207–17 e2. doi: 10.1053/j.gastro.2012.07.116.
342. Bluestone JA, Buckner JH, Fitch M, Gitelman SE, Gupta S, Hellerstein MK, Herold KC, Lares A, Lee MR, Li K, et al. Type 1 diabetes immunotherapy using polyclonal regulatory T cells. Sci Transl Med. 2015;7(315):315ra189. doi: 10.1126/scitranslmed.aad4134.
343. Canavan JB, Scotta C, Vossenkamper A, Goldberg R, Elder MJ, Shoval I, Marks E, Stolarczyk E, Lo JW, Powell N, et al. Developing in vitro expanded CD45RA+ regulatory T cells as an adoptive cell therapy for Crohn's disease. Gut. 2016;65(4):584–594. doi: 10.1136/gutjnl-2014-306919.
344. Moayyedi P, Surette MG, Kim PT, Libertucci J, Wolfe M, Onischi C, Armstrong D, Marshall JK, Kassam Z, Reinisch W, et al. Fecal microbiota transplantation induces remission in patients with active ulcerative colitis in a randomized controlled trial. Gastroenterology. 2015;149(1):102–109.e6. doi: 10.1053/j.gastro.2015.04.001.
345. Paramsothy S, Kamm MA, Kaakoush NO, Walsh AJ, van den Bogaerde J, Samuel D, Leong RWL, Connor S, Ng W, Paramsothy R, et al. Multidonor intensive faecal microbiota transplantation for active ulcerative colitis: a randomised placebo-controlled trial. The Lancet. 2017;389(10075):1218–1228. doi: 10.1016/S0140-6736(17)30182-4.
346. Cui B, Feng Q, Wang H, Wang M, Peng Z, Li P, Huang G, Liu Z, Wu P, Fan Z, et al. Fecal microbiota transplantation through mid-gut for refractory Crohn's disease: safety, feasibility, and efficacy trial results. J Of Gastro And Hepatol. 2015;30(1):51–58. doi: 10.1111/jgh.12727.
347. Shen J, Zuo ZX, Mao AP. Effect of probiotics on inducing remission and maintaining therapy in ulcerative colitis, Crohn's disease, and pouchitis. Inflammatory Bowel Dis. 2014;20(1):21–35. doi: 10.1097/01.MIB.0000437495.30052.be.
348. Oliva S, Di Nardo G, Ferrari F, Mallardo S, Rossi P, Patrizi G, Cucchiara S, Stronati L. Randomised clinical trial: the effectiveness of Lactobacillus reuteri ATCC 55730 rectal enema in children with active distal ulcerative colitis. Aliment Pharmacol Ther. 2012;35(3):327–334. doi: 10.1111/j.1365-2036.2011.04939.x.
349. Kim SO, Ah YM, Yu YM, Choi KH, Shin WG, Lee JY. Effects of probiotics for the treatment of atopic dermatitis: a meta-analysis of randomized controlled trials. Ann Allergy Asthma Immunol. 2014;113(2):217–226. doi: 10.1016/j.anai.2014.05.021.

350. Besselink MG, van Santvoort HC, Buskens E, Boermeester MA, van Goor H, Timmerman HM, Nieuwenhuijs VB, Bollen TL, van Ramshorst B, Witteman BJ, et al. Probiotic prophylaxis in predicted severe acute pancreatitis: a randomised, double-blind, placebo-controlled trial. The Lancet. 2008;371 (9613):651–659. doi: 10.1016/S0140-6736(08)60207-X.
351. Chambers ES, Viardot A, Psichas A, Morrison DJ, Murphy KG, Zac-Varghese SE, MacDougall K, Preston T, Tedford C, Finlayson GS, et al. Effects of targeted delivery of propionate to the human colon on appetite regulation, body weight maintenance and adiposity in overweight adults. Gut. 2015;64(11):1744–1754. doi: 10.1136/gutjnl-2014-307913.
352. Al KF, Craven LJ, Gibbons S, Parvathy SN, Wing AC, Graf C, Parham KA, Kerfoot SM, Wilcox H, Burton JP, et al. Fecal microbiota transplantation is safe and tolerable in patients with multiple sclerosis: a pilot randomized controlled trial. Mult Scler J Exp Transl Clin. 2022;8(2):20552173221086662. doi: 10.1177/20552173221086662.
353. Jacob V, Crawford C, Cohen-Mekelburg S, Viladomiu M, Putzel GG, Schneider Y, Chabouni F, O'neil S, Bosworth B, Woo V, et al. Single delivery of high-diversity fecal microbiota preparation by colonoscopy is safe and effective in increasing microbial diversity in active ulcerative colitis. Inflammatory Bowel Dis. 2017;23(6):903–911. doi: 10.1097/MIB.0000000000001132.
354. Arpaia N, Rudensky AY. Microbial metabolites control gut inflammatory responses. Proc Natl Acad Sci USA. 2014;111(6):2058–2059. doi: 10.1073/pnas.1323183111 .
355. Postler TS, Ghosh S. Understanding the holobiont: how microbial metabolites affect human health and shape the immune system. Cell Metab. 2017;26(1):110–130. doi: 10.1016/j.cmet.2017.05.008.
356. Isabella VM, Ha BN, Castillo MJ, Lubkowicz DJ, Rowe SE, Millet YA, Anderson CL, Li N, Fisher AB, West KA, et al. Development of a synthetic live bacterial therapeutic for the human metabolic disease phenylketonuria. Nat Biotechnol. 2018;36(9):857–864. doi: 10.1038/nbt.4222.
357. Berer K, Gerdes LA, Cekanaviciute E, Jia X, Xiao L, Xia Z, Liu C, Klotz L, Stauffer U, Baranzini SE, et al. Gut microbiota from multiple sclerosis patients enables spontaneous autoimmune encephalomyelitis in mice. Proc Natl Acad Sci USA. 2017;114(40):10719–10724. doi: 10.1073/pnas.1711233114.
358. Jangi S, Gandhi R, Cox LM, Li N, von Glehn F, Yan R, Patel B, Mazzola MA, Liu S, Glanz BL, et al. Alterations of the human gut microbiome in multiple sclerosis. Nat Commun. 2016;7(1):12015. doi: 10.1038/ncomms12015.
359. Sharon G, Cruz NJ, Kang DW, Gandal MJ, Wang B, Kim YM, Zink EM, Casey CP, Taylor BC, Lane CJ, et al. Human gut microbiota from autism spectrum disorder promote behavioral symptoms in mice. Cell. 2019;177 (6):1600–1618.e17. doi: 10.1016/j.cell.2019.05.004.
360. Gopalakrishnan V, Spencer CN, Nezi L, Reuben A, Andrews MC, Karpinets TV, Prieto PA, Vicente D, Hoffman K, Wei SC, et al. Gut microbiome modulates response to anti–PD-1 immunotherapy in melanoma patients. Science. 2018;359(6371):97–103. doi: 10.1126/science.aan4236.
361. Routy B, Le Chatelier E, Derosa L, Duong CPM, Alou MT, Daillere R, Fluckiger A, Messaoudene M, Rauber C, Roberti MP, et al. Gut microbiome influences efficacy of PD-1–based immunotherapy against epithelial tumors. Science. 2018;359(6371):91–97. doi: 10.1126/science.aan3706.
362. Blair HA. RBX2660 (REBYOTA®) in preventing recurrence of clostridioides difficile infection: a profile of its use in the USA. Drugs Ther Perspect. 2023;39(10):331–338. doi: 10.1007/s40267-023-01023-y.
363. Blair HA. SER-109 (VOWST™): a review in the prevention of recurrent clostridioides difficile infection. Drugs. 2024;84(3):329–336. doi: 10.1007/s40265-024-02006-7.
364. Petrof EO, Claud EC, Gloor GB, Allen-Vercoe E. Microbial ecosystems therapeutics: a new paradigm in medicine? Benef Microbes. 2013;4(1):53–65. doi: 10.3920/BM2012.0039.
365. O'Toole PW, Marchesi JR, Hill C. Next-generation probiotics: the spectrum from probiotics to live biotherapeutics. Nat Microbiol. 2017;2(5):17057. doi: 10.1038/nmicrobiol.2017.57.
366. Zmora N, Zilberman-Schapira G, Suez J, Mor U, Dori-Bachash M, Bashiardes S, Kotler E, Zur M, Regev-Lehavi D, Brik RBZ, et al. Personalized gut mucosal colonization resistance to empiric probiotics is associated with unique host and microbiome features. Cell. 2018;174(6):1388–1405.e21. doi: 10.1016/j.cell.2018.08.041.
367. Allegretti JR, Mullish BH, Kelly C, Fischer M. The evolution of the use of faecal microbiota transplantation and emerging therapeutic indications. Lancet. 2019;394 (10196):420–431. doi: 10.1016/S0140-6736(19)31266-8.
368. Maldonado-Gomez MX, Martinez I, Bottacini F, O'Callaghan A, Ventura M, van Sinderen D, Hillmann B, Vangay P, Knights D, Hutkins R, et al. Stable engraftment of bifidobacterium longum AH1206 in the human gut depends on individualized features of the resident microbiome. Cell Host & Microbe. 2016;20(4):515–526. doi: 10.1016/j.chom.2016.09.001.
369. Costello SP, Hughes PA, Waters O, Bryant RV, Vincent AD, Blatchford P, Katsikeros R, Makanyanga J, Campaniello MA, Mavrangelos C, et al. Effect of fecal microbiota transplantation on 8-week remission in patients with ulcerative colitis: a randomized clinical trial. JAMA. 2019;321(2):156–164. doi: 10.1001/jama.2018.20046.
370. Dsouza M, Menon R, Crossette E, Bhattarai SK, Schneider J, Kim YG, Reddy S, Caballero S, Felix C, Cornacchione L, et al. Colonization of the live biotherapeutic product VE303 and modulation of the microbiota and metabolites

in healthy volunteers. Cell Host & Microbe. 2022;30 (4):583–598.e8. doi: 10.1016/j.chom.2022.03.016.

371. Coyte KZ, Schluter J, Foster KR. The ecology of the microbiome: networks, competition, and stability. Science. 2015;350(6261):663–666. doi: 10.1126/science.aad2602.
372. Cao Z, Sugimura N, Burgermeister E, Ebert MP, Zuo T, Lan P. The gut virome: a new microbiome component in health and disease. EBioMedicine. 2022;81:104113. doi: 10.1016/j.ebiom.2022.104113.
373. Dalamaga M, Zheng L, Liu J. Gut mycobiome as a promising preventive and therapeutic target for metabolic disorders. Metabol Open. 2022;13:100168. doi: 10.1016/j.metop.2022.100168.
374. Scarpellini E, Ianiro G, Attili F, Bassanelli C, De Santis A, Gasbarrini A. The human gut microbiota and virome: potential therapeutic implications. Digestive And Liver Dis. 2015;47(12):1007–1012. doi: 10.1016/j.dld.2015.07.008.
375. Kashyap PC, Chia N, Nelson H, Segal E, Elinav E. Microbiome at the frontier of personalized medicine. Mayo Clinic Proc. 2017;92(12):1855–1864. doi: 10.1016/j.mayocp.2017.10.004.
376. Shaffer M, Armstrong AJS, Phelan VV, Reisdorph N, Lozupone CA. Microbiome and metabolome data integration provides insight into health and disease. Transl Res. 2017;189:51–64. doi: 10.1016/j.trsl.2017.07.001.
377. Xu J, Yang Y. Gut microbiome and its meta-omics perspectives: profound implications for cardiovascular diseases. Gut Microbes. 2021;13(1):1936379. doi: 10.1080/19490976.2021.1936379.
378. Geva-Zatorsky N, Sefik E, Kua L, Pasman L, Tan TG, Ortiz-Lopez A, Yanortsang TB, Yang L, Jupp R, Mathis D, et al. Mining the human gut microbiota for immunomodulatory organisms. Cell. 2017;168(5):928–943. e11. doi: 10.1016/j.cell.2017.01.022.