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Review

Integrative systems biology approaches for analyzing microbiome dysbiosis and species interactions

Syed Sabih Ur Rehman[1], Muhammad Ibtisam Nasar[1], Cristina S. Mesquita[2], Souhaila Al Khodor[3], Richard A. Notebaart[4], Sascha Ott[2,5], Sunil Mundra[1,6], Ramesh P. Arasardanam[5,7,8], Khalid Muhammad[1], Mohammad Tauqeer Alam[1,*]

[1]Department of Biology, United Arab Emirates University, Sheik Khalifa Bin Zayed St, Asharij, P.O. Box 15551, Al Ain, Abu Dhabi, United Arab Emirates
[2]Bioinformatics & Digital Health Services (RTP), University of Warwick, CV4 7AL, Coventry, United Kingdom
[3]Reproductive and Perinatal Health Division, Research Department, Sidra Medicine, Doha, Qatar
[4]Food Microbiology, Wageningen University and Research, 6700 AA, Wageningen, The Netherlands
[5]Warwick Medical School, University of Warwick, CV4 7AL, Coventry, United Kingdom
[6]Khalifa Center for Genetic Engineering and Biotechnology, United Arab Emirates University, Khalifa Bin Zayed Street, P.O. Box 15551, Al Ain, Abu Dhabi, United Arab Emirates
[7]Institute of Precision Diagnostics & Translational Medicine, University Hospitals Coventry & Warwickshire NHS Trust, Clifford Bridge Road, Coventry CV2 2DX, United Kingdom
[8]Leicester Cancer Research Centre, University of Leicester, LE1 7RH, Leicester, United Kingdom
*Corresponding author. Department of Biology, United Arab Emirates University, Sheik Khalifa Bin Zayed St, Asharij, P.O. Box 15551, Al Ain, Abu Dhabi, United Arab Emirates. Tel: +971-(0)3-7136370, E-mail: mtalam@uaeu.ac.ae

Abstract

Microbiomes are crucial for human health and well-being, with microbial dysbiosis being linked to various complex diseases. Therefore, understanding the structural and functional changes in the microbiome, along with the underlying mechanisms in disease conditions, is essential. In this review, we outline the structure and function of different human microbiomes and examine how changes in their composition may contribute to diseases. We highlight critical information associated with microbial dysbiosis and explore various therapeutic strategies for restoring a healthy microbiome, including microbiota transplantation, phage therapy, probiotics, prebiotics, dietary interventions, and drug-based approaches. Further, to better understand microbiome dysbiosis, we discuss multi-omics approaches including metagenomics, metatranscriptomics, metaproteomics, and meta-metabolomics, alongside computational modeling approaches such as ecological and metabolic network analysis. We outline key challenges associated with multi-omics techniques and emphasize the importance of integrative systems biology approaches that combine multi-omics data with computational modeling. These approaches are crucial for effectively analyzing microbiome data, providing deeper insights into species interactions and microbiome dynamics. Finally, we offer insights into future research directions in the field of microbiome research. This review makes a unique contribution to microbiome research by presenting a holistic framework that integrates multi-omics data with multi-scale modeling to elucidate microbial interactions, microbiome dysbiosis, and their modulation in disease-associated contexts.

Keywords: microbiomes; microbial dysbiosis; metagenomics; gut microbiome; microbiome-associated disease; mathematical modeling; microbial community modeling; ecological network model; genome-scale metabolic model; integrative systems biology

Introduction

Microbial dysbiosis, a disruption in the microbial ecosystem, has been associated with a wide range of diseases, including metabolic, inflammatory, neurological, hepatic, and oncological conditions, impacting multiple organ systems throughout the body [1]. In these disease conditions, the abundance of beneficial microbes is reduced, and the pathogenic microbes amplify [2, 3]. This pathogenic shift of microbes leads to complexity in microbiome functionality and results in disease phenotypes. In most cases, microbial dysbiosis is caused by various factors, including diet, environment, aging, drug consumption, and disease status [4–9]. Moreover, certain bacterial families such as *Lachnospiraceae, Erysipelotrichaceae, Veillonellaceae, Enterobacteriaceae* (*Proteobacteria*), and many others may be linked to metabolic impairments, potentially contributing to adverse health effects [10–12]. In contrast, bacterial families such as *Bacteroidaceae, Prevotellaceae, Bifidobacteriaceae*, or *Clostridiaceae*, among many others, are associated with healthier metabolic states, indicating a more normal and functional microbiome [10]. These beneficial microbes often engage in mutualistic relationships with their host, aiding in essential processes like digestion [13], immune system modulation, and protection against pathogenic organisms [14]. Some of the species of these families are associated with polysaccharide breakdown, short-chain fatty acid (SCFA) production, fermentation of fiber, and butyrate production [13]. Understanding the dynamics of microorganisms within the microbiome is crucial for gaining deeper insights into disease mechanisms. Recent advancements in high-throughput sequencing technologies and systems biology approaches, particularly genome-scale metabolic modeling [15] integrated with multi-omics

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datasets, have significantly improved our understanding of microbiome complexity. These developments have provided deeper insights into compositional and functional microbial shifts caused by dysregulation and dysbiosis, highlighting their integral role in human health [16].

In this review, we highlight the key role of systems biology approaches in advancing microbiome research by using computational modeling and simulation to understand complex microbial data. We explore various computational models, including network-based models that map and analyze interactions between microbial species, and genome-scale metabolic models that reconstruct metabolic networks from genomic data to predict the functional capabilities of individual microbes or entire communities. Further, by integrating these computational models with multi-omics datasets, this review highlights their potential in explaining microbial community dynamics, disease mechanisms, and potential therapeutic interventions. We also discuss challenges like data quality and computational limits, offering insights into future solutions. This review provides a comprehensive overview, emphasizing a multi-scale approach that will benefit researchers aiming to use systems biology to fully explore microbiome studies, ultimately leading to health and disease management advances.

Microbiome structure and composition

The number of microbial cells in the human microbiome is nearly equal to human cells, totaling $\sim 3.8 \times 10^{13}$ bacterial cells and 3×10^{13} human cells [17]. The human microbiome exhibits remarkable genetic diversity with ~46 million nonredundant microbial genes (~22 million in the gut and ~24 million in the oral microbiome), vastly exceeding the ~23 000 protein-coding genes in the human genome [18]. These findings show the microbiome's potential role in shaping individual health and phenotypic diversity. The human microbiome comprises a complex community of microorganisms, including bacteria, archaea, viruses, and eukaryotic microbes such as fungi and protozoa [19, 20]. The bacteria predominate within the microbiome; however, they contribute significantly less by weight, which is ~200 g of body mass [17]. Microbiome is initially acquired at birth, primarily through exposure to the mother's vaginal and skin microbiota, along with early environmental contact. It then develops into a complex and dynamic ecosystem, mainly influenced by factors such as diet, environment, antibiotic exposure, and health status [21, 22].

Microorganisms inhabit various body sites, including the skin, oral cavity, respiratory tract, urogenital tract, and gastrointestinal tract, each hosting unique microbial communities, playing vital roles in human well-being [23]. For example, the skin microbiome protects against pathogens, modulates immune responses, and contributes to skin health [24]. Oral health is maintained by the oral microbiome, which initiates digestion and prevents colonization by pathogens [18]. Respiratory microbiome influences respiratory health and immune responses, and any disruption of microbial structure can lead to conditions like asthma and chronic obstructive pulmonary disease (COPD) [25, 26]. For instance, an increase in Proteobacteria genera such as *Haemophilus influenzae* and *Moraxella catarrhalis* is associated with asthma and COPD [27]. COPD patients also exhibit a decrease in the relative abundance of *Prevotella* and *Veillonella* [28, 29]. In the female urogenital microbiome, the vaginal microbiome is crucial for reproductive health, preventing infections, maintaining pH balance [30], and its composition and diversity have also been associated with preterm birth [31]. Similarly, the gut microbiome is central to various essential biological processes such as maintaining homeostasis [32], nutrient absorption [33], immune system maturation [14], and resistance to pathogens [34]. The microbiota plays crucial roles in the gut–brain axis through neurological, immunological, and endocrine pathways, affecting neurotransmitters like serotonin, dopamine, and gamma-aminobutyric acid, thereby impacting mood, cognition, and learning [35, 36]. It also regulates the hypothalamic–pituitary–adrenal axis, the body's primary stress response mechanism, and influences the circadian rhythm, governing sleep–wake cycles [37, 38]. Recently, a study identified a distinct brain microbiome in healthy *Oncorhynchus mykiss* (rainbow trout) and *Salmo salar* (Atlantic salmon), suggesting that microbiota-driven neuromodulation occurs directly in the brain through symbiotic microbial presence influencing vertebrate neural systems, even under physiological (nondisease) conditions [39, 40].

Among all body sites, the human gut hosts the largest and most diverse microbiome, primarily composed of bacteria from four phyla: *Firmicutes*, *Bacteroidetes*, *Actinobacteria*, and *Proteobacteria*, with *Proteobacteria* being the largest [41]. Each of these microbial groups plays a distinct role in maintaining human health [42]. For example, *Firmicutes* members like *Faecalibacterium prausnitzii* and *Christensenella minuta* in immune modulation and *Roseburia* produce butyrate, a short-chain fatty acid (SCFA) that serves as an energy source for colonocytes, promotes anti-inflammatory responses, and strengthens gut barrier function, establishing a healthy community structure [42–47]. Metabolically active *Firmicutes, Bacteroidetes,* and *Verrucomicrobia* phyla are involved in the breakdown of complex carbohydrates, protein metabolism, and production of SCFAs like acetate and propionate, influencing energy homeostasis and lipid metabolism [48, 49]. *Actinobacteria* include *Bifidobacterium* species, which are important for fermenting dietary fibers and producing vitamins such as vitamin B12 [50, 51], vitamin K [52], and also regulate vitamin D [53]. Moreover, species of *Proteobacteria*, on the other hand, participate in nitrogen fixation, supporting gut homeostasis and various other metabolic processes, emphasizing the significant role for sustained well-being. Gut microbiome plasticity, as well as variability in the structure and composition, is crucial for maintaining host health [42]. This person-specific microbiome contributes to the uniqueness of each individual's microbial ecosystem and could play a role in developing personalized medicines. For instance, microbial profiling has been used to predict dietary response and immunotherapy outcomes, with *Akkermansia muciniphila* abundance linked to enhanced anti-PD-1 efficacy [51, 54]. However, its vast diversity presents challenges in studying the microbiome collectively, making it difficult to identify universal patterns [55–57]. Therefore, large, well-controlled studies are essential to draw meaningful conclusions about microbiome-related health and disease.

Microbiome dysbiosis and its implication in diseases

A state of balance and health in the microbial communities, referred to as eubiosis, supports host physiological homeostasis. It enhances protection against pathogens and promotes resilience to environmental and physiological challenges [58, 59]. Disruption of microbial diversity, on the other hand, can lead to a state of imbalance known as dysbiosis [7, 60]. Microbiome dysbiosis refers to a deviation from the usual microbial composition observed in healthy hosts. However, there is no clear accepted definition of

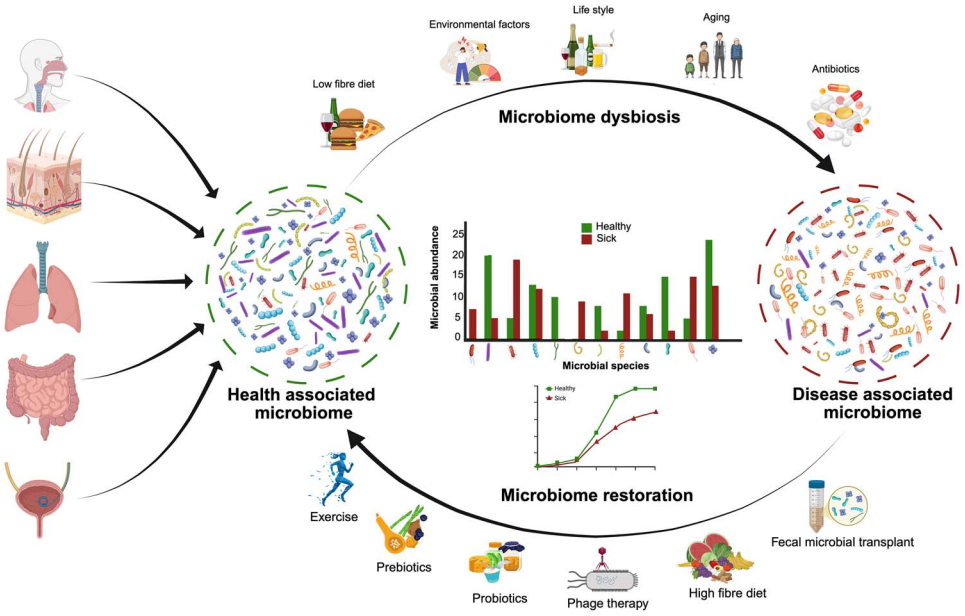

Figure 1. Human microbiome dysbiosis and restoration. A comprehensive overview illustrating various factors associated with microbiome dysbiosis leading to a reduction in microbial diversity, a decline in beneficial taxa, and an overrepresentation of pathogenic microorganisms, contributing to disease progression. Potential therapeutic interventions have been explored for microbiome restoration and disease mitigation.

healthy microbiome structure, and it can vary between individuals. It is often marked by a loss of beneficial microbes, overgrowth of harmful organisms, or reduced microbial diversity, with various external factors regulating microbiome states [4]. Some of the key contributors to microbiome dysbiosis include diet, environment, age, antibiotics, immunity, and lifestyle (Fig. 1). Among these, diet is considered one of the most significant contributors, with fiber-rich diets promoting microbial diversity and supporting health, while high-fat and high-sugar diets reduce diversity and drive dysbiosis by increasing bile-tolerant bacteria like *Alistipes*, *Bilophila*, and *Bacteroides* [61–63]. Similarly, antibiotic use can severely disrupt the microbial structure by eradicating beneficial bacteria alongside pathogens, causing long-term microbiome alterations [6, 64]. Aging is also linked to microbiome shifts that reduce diversity and alter immune function [5, 65], while immune deficiencies or overactivation can disrupt microbial equilibrium, contributing to conditions like inflammatory bowel disease (IBD) and allergies [66]. Lifestyle factors, including stress, physical activity, and sleep patterns, also shape gut microbiota [67–70], while environmental exposures, geographical location, pollution, and toxins further influence microbial communities [9, 71, 72].

Microbiome dysbiosis has been linked to a large number of complex human diseases including different gastrointestinal conditions [73], metabolic disorders [74], cardiovascular diseases [75], neurological disorders [76, 77], liver conditions [78], autoimmune and inflammatory diseases [79], skin conditions [80], and cancers [81]. One of the gastrointestinal conditions is IBD, which includes Crohn's disease (CD) and ulcerative colitis (UC), which are closely associated with gut microbiome dysbiosis [82]. These conditions are characterized by reduced microbial diversity, instability, and an increase in pro-inflammatory bacteria, such as *Enterobacteriaceae*, alongside a decline in anti-inflammatory species like *F. prausnitzii* and *Roseburia* [83]. This dysbiosis drives intestinal inflammation, barrier dysfunction, and immune dysregulation [84]. Similarly, colorectal cancer, a leading global cause of mortality, dysbiosis manifests as an overabundance of *Escherichia coli* and *Bacteroides fragilis*, which is reported to promote chronic inflammation and tumorigenesis [85–87]. Moreover, the gut microbiome composition of pathogenic microbes has long been emerging as a potential biomarker for predicting colon cancer recurrence [88–91].

Similarly, studies have highlighted a reduced abundance of *Firmicutes* and an increase in *Bacteroidetes* in obesity and type 2 diabetes (T2D), contributing to metabolic dysfunction through reduced SFCA production and impaired gut barrier integrity, causing inflammation [92]. Gut microbiome dysbiosis may influence lipid metabolism and inflammation that have been implicated in cardiovascular diseases such as atherosclerosis and hypertension [75, 93–96]. In autism, changes in gut microbiota composition, such as decreased *Bifidobacterium* and increased *Clostridium*, and reduced bile acids and lipids are believed to impact brain development and behavior [97, 98]. Several studies suggest a profound connection between gut microbial health and neurodevelopmental outcomes, such as modulation of microglial maturation, blood–brain barrier integrity, and neurotransmitter signaling [77, 98–101]. The gut community structure can influence blood–brain barrier via SFCAs like butyrate, which enhance tight junction expression, and modulates neuroinflammation by regulating systemic immune responses [99, 101]. Studies reveal that microbial patterns, such as those observed in Parkinson's disease, are influenced by medication and precede neurological symptoms, suggesting early gut dysregulation in the onset and progression of neurodegenerative diseases [102, 103]. Conversely, in a healthy state, microbial metabolites such as SCFAs, Indole-3-propionic acid (IPA), and Trimethylamine-N-oxide (TMAO) act as systemic signaling molecules that strengthen epithelial barriers (tight junction) and modulate inflammation, protecting against chronic disease across the gut–brain–liver axis [104–106].

Microbial dysbiosis during infancy and early childhood, affecting both the gut and lungs, can predispose individuals to asthma by disrupting immune regulation and promoting airway inflammation [107]. Particularly, the dominance of pathogenic bacteria like *Haemophilus* and *Moraxella*, in the lung has been associated with the persistence and severity of asthma [26, 108].

This dysbiosis disrupts the gut–lung axis, a bi-directional interaction between the gut and lung microbiomes, leading to chronic inflammation and allergic responses in the respiratory system [109, 110]. Autoimmune disorders such as inflammatory arthritis, including rheumatoid arthritis (RA), are linked to dysbiosis caused by enrichment of pro-inflammatory and oral taxa *Ruminococcus gnavus*, *E. coli*, *Streptococcus* spp., and depletion in SFCA-producing bacteria, causing functional shifts in metabolism and stress pathways [111], while early onset of RA is linked to the enrichment of *Prevotella copri* in patients [79]. Skin diseases such as atopic dermatitis (AD) and psoriasis are also influenced by gut microbiome dysbiosis [112]. The skin acts as a physical, microbial, chemical, and immune barrier, and the skin microbiome modulates immunity and other barrier sites in the body [113, 114], which involves the dominance of *Fusobacterium, Trueperella, Staphylococcus, Streptococcus*, and *Bacteroides* and alterations in metabolites like tryptophan, which impact skin barrier function and immune activation [80, 115].

Restoring the healthy state of the human microbiome

Restoring a healthy microbiome in patients has been shown to reduce disease symptoms and improve overall health [116]. Over the years, various approaches for microbiome engineering in restoring a healthy microbiome state have proven successful. Some of the most effective methods include fecal microbiome transplant (FMT) [117], probiotics [118], prebiotics [119], diet [63], drugs [120], phage therapy [121], and exercise [122] (Fig. 1).

FMT involves transferring stool from healthy donors to a patient's, is among the most effective methods for increasing microbial diversity and has been used to treat various conditions, including IBD, irritable bowel syndrome [117], obesity, antibiotic resistance, inflammation, constipation, food allergy, and recurrent *Clostridioides difficile* colitis [123, 124]. Treating early-stage Parkinson's disease (PD) patients, rebalancing immunity, and suppressing atopic dermatitis allergic response with FMT have shown beneficial effects on symptoms and demonstrated the positive modulation of the gut microbiome as a potential therapeutic approach [125, 126]. However, the limitations of FMT include certain risk factors such as variable composition of donor microbiota, lack of standardized protocols, and inconsistent long-term efficacy and transmission of resistant bacteria from donor to patient [127, 128]. Therefore, it is not yet a routine therapy. To improve therapeutic efficacy, efforts include standardizing donor screening, developing defined microbial consortia, implementing metagenomic testing [129–131], and advancing Food and Drug Administration (FDA)-approved therapies such as RBX2660 (Rebyota) and SER-109 (Vowst) for recurrent *C. difficile* infection [132, 133].

Diet and prebiotics also play an important role in modulating gut health. Prebiotics such as inulin, glucan, fructans, galacto-oligosaccharides, or short polysaccharides can pass through the gastrointestinal tract undigested and help in the growth of beneficial gut microbial genera [134] such as *Bifidobacterium* and *Lactobacillus* through selective fermentation and synergistic interaction, supporting each other's colonization and metabolic activity [134]. Certain diets like the fiber-rich Mediterranean diet, low in saturated fats, affect the composition and function of the gut microbiota and influence metabolic homeostasis and overall well-being [63]. It increases the abundance of beneficial taxa like *F. prausnitzii* and *A. muciniphila*, which support gut barrier integrity, enhance SCFA production, and reduce systemic inflammation, a key driver for maintaining metabolic homeostasis [63]. Specific dietary components, hippurate derivative metabolites from coffee, increase beneficial microbes like *Lawsonibacter asaccharolyticus* by serving as selective metabolic substrates that promote competitive fitness of the gut microbial community [135]. Interventions such as intermittent fasting [136], reduced-calorie diets [137], fenugreek supplementation [138], fiber-rich diet [139], and nicotinamide *N*-methyltransferase inhibition [140], significantly modulate the gut microbiome, promoting beneficial microbial populations, enhancing diversity, and improving metabolic outcomes [8]. Moreover, synergistic effects such as combining a fiber-rich diet with prebiotics (such as inulin) have shown greater enhancement of beneficial taxa and SCFA production than either intervention alone [8].

Several drugs have demonstrated some beneficial effects on the microbiome. For instance, metformin interacts with the gut microbiome, potentially by enhancing mucin layer integrity and SCFA production, thereby altering microbial composition in treatment-naive patients with T2D [120]. After treatment with metformin, the abundance of *A. muciniphila*, *E. coli*, *Bifidobacterium* spp., and *Blautia wexlera* increased, promoting anti-inflammatory effects and improved insulin sensitivity. However, it can also reduce the abundance of some anti-inflammatory species [141]. Various other compounds such as SCFAs, polyphenols, omega-3 fatty acids, berberine, and glucagon-like peptide-1 (GLP-1) receptor agonists have shown significant potential in positively modulating the gut microbiota [142–144], offering therapeutic benefits in microbiome-related diseases [121].

Phage therapy, commonly used as an antimicrobial treatment, can also modulate the structure and composition of the microbiome [121]. In a recent study, it was shown that phage therapy can selectively target gut *Fusobacterium nucleatum* (Fn), a species that impairs the host's anticancer immune response by promoting the expansion of immunosuppressive myeloid-derived suppressor cells (MDSCs) in the gut. In particular, the study demonstrated that an *Fn*-binding M13 phage is capable of targeting *Fn* in the gut. This results in a reduction of MDSC amplification at the tumor site, potentially enhancing the host's anticancer immune response [66]. Phage therapy and microbiota-based treatments offer promising alternatives for antibiotic-resistant and chronic bacterial infections, demonstrating clinical efficacy in 'treating *Mycobacterium*-related infections' [145], *Pseudomonas aeruginosa* [146, 147], and *C. difficile* infections [148]. While phage–antibiotic synergy to restore microbiome shows potential for long-term therapeutic success [149, 150], with a limited number of phages and variable responses in patients, it requires optimization of the protocol for consistent results [3].

The use of probiotics also positively modulates microbiome dysbiosis. Probiotics are beneficial living microorganisms that often compete with the group of harmful microbes in the gut or sometimes produce antimicrobial substances that can directly kill harmful bacteria. As a result, probiotics increase microbiome diversity [151]. A combination of various gut microorganisms, such as *Lactobacillus*, *Bifidobacteria*, and yeasts, can be taken in prescribed doses to improve health conditions [152]. Several studies have demonstrated the use of probiotics and prebiotics to treat dysbiosis [153], including engineered *Saccharomyces cerevisiae* for IBD [154], and *E. coli* probiotics [155], demonstrating a targeted approach [156] in managing conditions such as IBD, melanoma, and Alzheimer's [118, 157–159].

While drugs can influence the microbiome, they may have broad or unintended effects, potentially disrupting microbial diversity by decreasing beneficial microbes [6, 160, 161] and causing side effects [141, 162]. On the other hand, microbial

modulation using probiotics, FMT, or phage therapy offers a more targeted approach to restore healthy gut microbial composition [163]. Probiotic-based therapies are generally considered safer but less potent than FMT or phage therapy, which offer broader microbiome modulation but carry higher risks and regulatory complexity. Please refer to Zitvogel *et al.* [164] for more details.

Analyzing microbiome structure and dysbiosis using multi-omics approaches

To effectively manage disease conditions by positively modulating microbiome structure requires a systems-level understanding of microbiome dynamics [165–167] (Table 1). This also includes understanding how the microbiome structure changes in disease settings and how these changes are linked to disease phenotypes. Various experimental and computational approaches have been successfully developed to study microbiome dysbiosis and its associated functional complexities across diverse conditions. There is a range of omics techniques including amplicon sequencing, whole genome metagenomic sequencing, single-cell sequencing, metatranscriptome, metaproteome, and meta-metabolomics, as well as computational modeling approaches [168–170] (Table 1, Fig. 2).

Amplicon sequencing is a DNA sequencing method that targets specific genomic regions, such as the 16S ribosomal RNA (rRNA) gene for bacteria or 18S for eukaryotes such as protists and microalgae, and Internal Transcribed Spacer (ITS) regions for fungi. This culture-independent approach is widely used to study species identification, microbial diversity, and taxonomic profiling across various environments [41, 171] (Table 1). Its major advantages include being cost-effective, relatively easy to analyze, and effective in identifying microorganisms that cannot be cultured in the lab [172, 173]. However, it has limitations, such as providing no information about functional genes or nontargeted organisms such as viruses, and often not resolving closely related species or strains [169, 174]. Additionally, amplicon sequencing data provide a probabilistic snapshot of microbial communities rather than a direct count, and biases can arise due to variations in rRNA gene copy numbers among taxa. To better approximate true community structures, it is important to normalize sequence counts based on gene copy numbers or to focus on presence/absence data instead of relative abundances [175, 176]. Nevertheless, whole-genome metagenomic sequencing is a comprehensive approach that sequences all the genetic material in a sample and identifies microbial community structures (Table 1); it provides valuable insights into functional genes and interactions within microbial communities. Moreover, this method captures the entire genomic content, including bacteria, archaea, fungi, viruses, and other microorganisms, and can differentiate closely related species or strains within a community. However, it is more expensive and involves intensive data analysis compared to amplicon-based sequencing [167, 169, 177].

Single-cell sequencing has transformed microbiome research by isolating microbes individually at high resolution. It overcomes the limitation of bulk multi-omics by discovering rare taxa, providing insights into functional heterogeneity and spatial organization in microbial ecosystems [178]. Single-cell strategies have enhanced our understanding by identifying dietary fiber responders driving metabolic shifts [179], and with synthetic biology tools, enabled precise strain-level tracking and functional profiling [180, 181]. In complex ecosystems, single-cell transcriptomics identifies metabolic niche differentiation across thousands of microbial species and reveals functional specialization, allowing the identification of metabolic stability and adaptation in a community [182]. Primarily, this technique holds promise for advancing microbiome-based therapeutics by overcoming bulk sequencing limits through single-cell resolution to enable precision diagnostics and personalized medicine [183]. Although it struggles with throughput due to low scalability, as it allows for only a limited number of microbes to be analyzed simultaneously, making large microbiome studies challenging [178].

The metatranscriptome approach involves sequencing the expressed RNA from all microorganisms within a microbiome (Table 1). Unlike whole-genome metagenomic sequencing, it focuses on active genes being expressed rather than just the genes present, offering insights into the metabolic activities and functional potential of microbial communities [169]. Similarly, the metaproteome approach gives insights into the expressed proteins from different microorganisms within the microbiome in disease and healthy states [184] (Table 1). It also provides a deeper understanding of the biological functions, including metabolism, signaling, and other biological processes within the microbiome; however, low-abundance proteins are difficult to detect in mass spectrometry [185, 186] (Table 1). Metametabolomics is used to identify the entire set of metabolites produced within the microbiome and to differentiate the chemical composition between microbial communities [187] (Table 1).

These multi-omics approaches are used to distinguish functional differences between microbiomes [177, 188–190]; however, techniques such as metatranscriptomics, metaproteomics, and metametabolomics remain less common due to their greater complexity and sensitivity compared to amplicon-based and metagenomic methods [169, 189] (Table 1). Furthermore, microbiome data analysis is often complicated by characteristics such as zero inflation, overdispersion, compositionality, and sample heterogeneity, which require specialized statistical approaches to reduce bias. When multiple of these features are present, more sophisticated and customized analytical frameworks become necessary [191–193]. Cumulatively, traditional multi-omics approaches provide valuable data but are unable to predict dynamic microbial behavior or reveal the mechanistic understanding of microbial interactions at the system level [194].

Ecological network models to understand microbiome ecosystem dynamics

Computational models of microbial communities serve as powerful tools for analyzing microbial interactions and microbiome dynamics to identify key species and metabolites [111]. They offer insights into community structure, dysbiosis, microbiome modulation, and the functional capabilities of individual microorganisms and the entire microbial ecosystem [233–236]. 'Ecological' network modeling (ENM) is one of the most widely used methods for understanding complex microbial interactions [237]. In this modeling approach, microbial communities are represented as networks, where nodes represent microbial species (or taxa) and edges indicate relationships. Based on their effects on participating microbes, microbial interactions within an ecosystem can be categorized into various relationship types, including mutualism (+/+), competition (−/−), parasitism (+/−), predation (+/−), commensalism (+/0), and amensalism (−/0) [42, 88] (Fig. 3).

Mutualistic interactions occur when both microorganisms benefit, often through cross-feeding (Fig. 3) [238]. For example, mutualistic interactions between *Bacteroides* and methanogens, a type of archaea, where *Bacteroides* break down complex carbohydrates into SCFAs and hydrogen, while methanogens use the hydrogen to produce methane, keeping the fermentation environment

Table 1. Comparative overview of multi-omics strategies in microbial community analysis.

Omics domain	Analytical goal	Use case	Key tools/platforms	Strengths	Limitations
Amplicon Sequencing	16S/ITS rRNA analysis, taxonomic profiling, alpha/beta diversity	Microbial diversity tracking in dysbiosis or environmental samples	QIIME2 [195], DADA2 [196], mothur [197]	Cost-effective. Sensitive to low-abundance taxa	PCR bias (i.e. primer specific), limited resolution with no functional insight.
Whole-Genome Metagenomics	Assembly, binning, taxonomy, functional metagenomics	Novel gene/pathway discovery in gut microbiome or soil studies	MEGAHIT [198], MetaSPAdes [199], MetaBAT2 [200], CheckM [201], metaQUAST [202], DRAM [203], metaFlye [204], HiFi-MAGs [205] Kraken2 [206], Metaphlan [207], HUMAnN [208]	Whole genome resolution, functional potential insights, novel species/pathway discovery.	Complex assembly in highly diverse communities, contamination in bins, and high-performance computation required
Metatranscriptome	Gene expression, active pathways analysis,	Identifying active genes during infection or treatment response	Salmon [209], Kallisto [210], DESeq2 [211], edgeR [212]	Functional insight into active metabolism	RNA instability, host/rRNA contamination, limited functional annotation for uncultured taxa.
Metaproteome	Protein identification, peptide quantification, functional annotation	Quantifying enzyme profiles in disease versus control samples	MaxQuant [213], Proteome Discoverer [214], MetaProteomeAnalyzer [215], MSFragger [216], Perseus [217], MSstats [218], metaQuantome [219], MaxDIA [220] DIA-NN [221]	Functional insight into enzyme activity, PTM (post-translational modifications) detection	Dynamic range of peptide quantification, database dependent, low protein yield
Metametabolomics	Metabolite identification, spectral matching, pathway enrichment	Detecting metabolite shifts in dietary intervention studies	MZmine 3 [222], XCMS [223], GNPS [224], MetFrag [225], MS-FINDER [226], Mummichog [227].	Real-time metabolic readout, identifies biomarkers, integration with other OMICS data	High sample variability, an extensive spectral library is required, metabolite identification ambiguity
Multi-Omics Integration	Correlation of microbial profiles with host or functions	Linking microbial shifts to host phenotypes or treatment outcomes	MixOmics [228], MOFA [229], MetaboAnalyst [230], OmicsNet [231], OpenMS3 [232]	Integrating across datasets reveals cross-talk mechanisms	Data normalization and interpretation complexity

for *Bacteroides* efficient [239]. Similarly, metabolic crossfeeding enhances the survival of mutually interacting microbes [240], and cooperating microbes tolerate antimicrobial drugs compared to noninteracting species [241]. In contrast, competitive interactions arise when microbes compete for limited resources (Fig. 3), often leading to mutual inhibition [242, 243]. For example, in the gut, *Lactobacillus* competes with *C. difficile* for shared common nutrients, keeping *C. difficile* in check and limiting its colonization. However, when *Lactobacillus* levels decline due to dysbiosis, *C. difficile* gains a competitive advantage, increasing infection risk. Pathogens like *Salmonella Typhimurium* and *P. aeruginosa* outcompete other microbes in inflammation by producing targeted toxins and biofilms that selectively target *Enterobacteriaceae*, influencing disease progression [244, 245]. In parasitism, one organism benefits while harming another (Fig. 3). For example, *Mycoplasma* species parasitize *Staphylococcus aureus* by extracting essential metabolites like amino acids and nucleotides, weakening *S. aureus* and increasing its susceptibility to environmental stress and immune defences. Indeed, predation interaction involves one organism actively preying on another (Fig. 3). For instance, *Bdellovibrio bacteriovorus* is a predatory bacterium that infects and kills *E. coli*, but *E. coli* does not mount a counter-infection or immune response. In commensalism, one species benefits without affecting the other (Fig. 3), for example, the interaction between *Lactobacillus* and *Bifidobacterium* in the gut, where *Lactobacillus* creates an acidic environment that promotes its own survival and prevents the growth of pathogens, which incidentally also supports *Bifidobacterium*, which thrives in the same conditions. Lastly, in amensalism, one organism is harmed while the other remains unaffected (Fig. 3). For example, the interaction between *Streptomyces* species and *E. coli*, where *Streptomyces* produces antibiotics that kill *E. coli*; however, *E. coli* does not affect *Streptomyces* in return. Each of these interactions plays a role in shaping microbial community dynamics and is closely linked to dysbiosis [235, 246, 247].

ENMs are developed using a combination of multi-omics datasets and various statistical and mathematical techniques such as differential equations and graph theory (Fig. 3). The

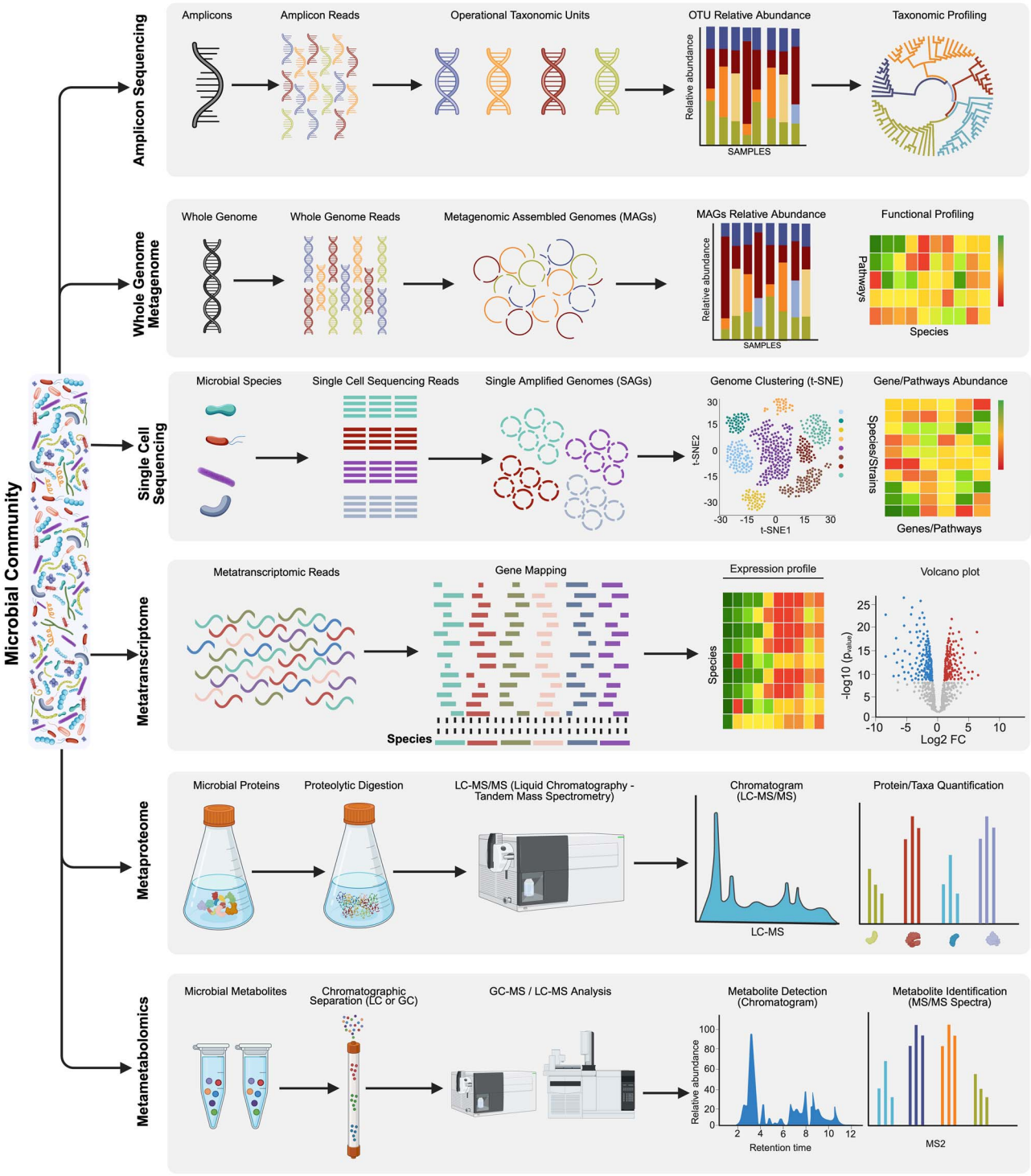

Figure 2. Multi-omics approaches to investigate microbiome characterization. This figure illustrates the main steps of multi-omics approaches used to study microbiome composition, function, and dynamics. Amplicon sequencing profiles microbial communities based on marker genes, taxonomic classification, relative abundance estimation, and phylogenetic tree construction. Whole-genome metagenomics assembles microbial genomes, facilitates the study of microbial diversity, analyzes genetic composition, and investigates metabolic pathways. Single-cell genomics isolates individual microbial cells, followed by whole-genome amplification and sequencing, enabling the detailed genetic characterization of microorganisms. Metatranscriptomics captures actively expressed genes across microbial communities, providing insights into metabolic activity and differentially expressed genes across conditions. Similarly, metaproteomics studies protein abundance, enzymatic activity, and microbial functional potential, and metabolomics detects and quantifies metabolites, enabling the characterization of metabolic signatures and biochemical pathways within microbial communities.

main approaches in ENM include co-occurrence network models, agent-based models (ABMs), cluster-based network models, dynamic models, temporal and spatial network models, and host-mediated microbiome interaction models, among others. Co-occurrence models rely on correlation patterns in species abundance within microbial communities [248–251]. However, their outcomes are highly sensitive to methodological choices, as described in the practical guide by Liu *et al.* [252]. Several

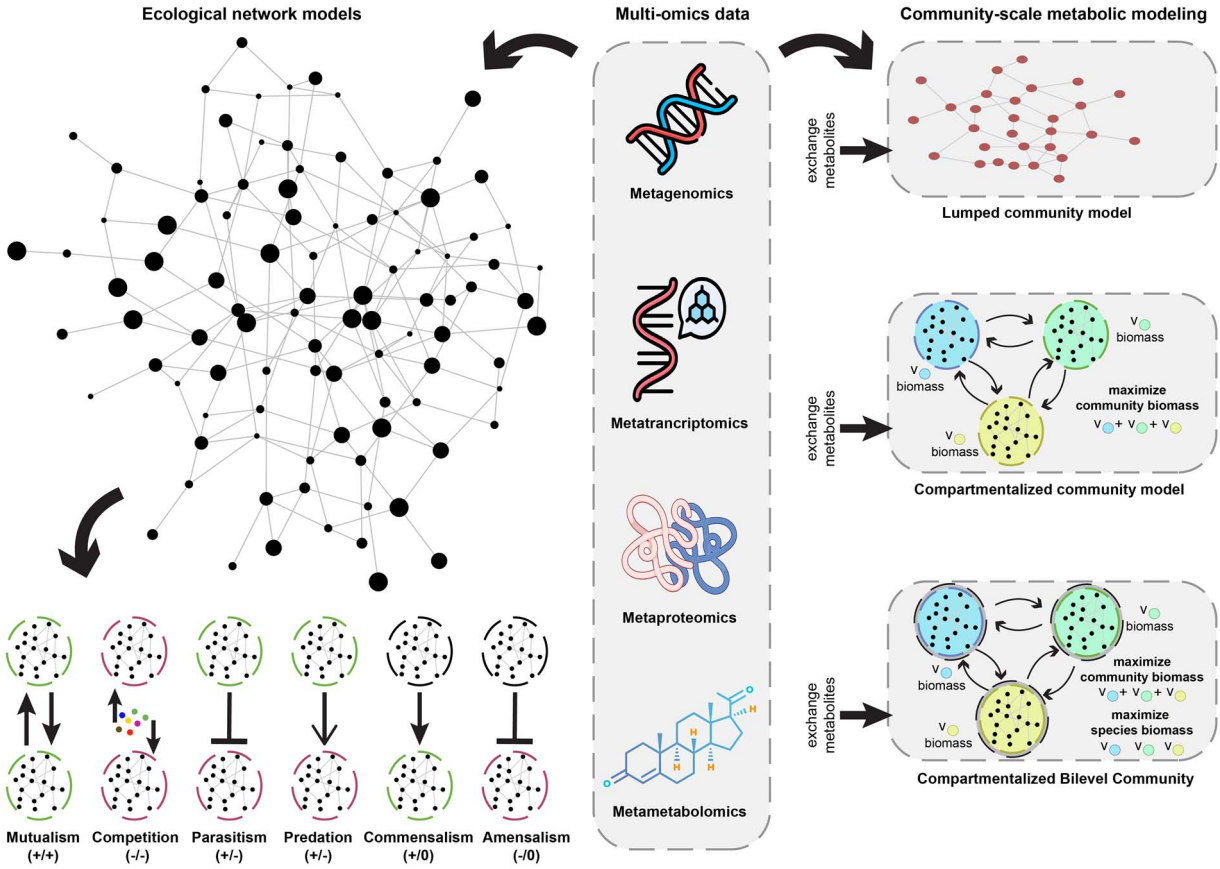

Figure 3. Integrative systems biology approach for reconstructing and characterizing ecological and metabolic networks of microbiomes. Multi-omics datasets, integrated with mathematical models, are used to reconstruct ecological and various types of metabolic networks in microbiomes, including lumped, compartmentalized, and nested community models. These network models facilitate system-level understanding of microbiomes, providing insights into microbial interactions, the metabolic capabilities of individual microorganisms, and the overall metabolic profiles of microbial communities.

microbial species co-occur in the human gut and have a positive correlation. For example, in *E. coli* and *Enterococcus*, both facultative anaerobes, and *Bacteroidetes* and *Firmicutes,* metabolize carbohydrates that coexist in the gut. This is evident in co-occurrence networks, which infer statistical correlations but not causation, and often oversimplify microbial relationships by assuming binary interactions (presence/absence), overlooking quantitative dynamics and limiting the prediction of external effects on dysbiosis, lacking mechanistic resolution [251, 253]. ABMs simulate individual microbial dynamics and interactions, providing a bottom–up approach to microbiome modeling [254], while cluster-based network modeling identifies functionally related microbial groups by examining modular structures within microbial networks and captures nonlinear dynamics among species without prior assumptions [255]. Dynamic modeling, on the other hand, relies on nonlinear differential equations such as generalized Lotka–Volterra equations to describe the population dynamics of different microbial species over time [256, 257]. The dynamic modeling approach is useful in understanding microbial ecosystem stability.

Moreover, as we know, the microbial community structure changes over time and space, so it is crucial to understand the microbial population dynamics over time and space. Longitudinal gut microbiome studies are based on temporal models and test how a certain diet or disease status changes gut microbiome dynamics over time. For instance, ECN (Ecological Normal Modes) modeling approach analyses microbiome fluctuation and stability by decomposing time series data into interpretable dynamic modes [258], while LUPINE is a regression-based framework that infers dynamic microbial interaction networks across time by using information from past time points to capture evolving inter-taxa relationships [259]. Similarly, spatial modeling approaches examine how species interactions vary across different locations in an ecosystem. These comprise joint species distribution models (JSDMs), with spatially structured latent variables for modeling environmental gradients and species associations [260]; Hierarchical Modelling of Species Communities (HMSC) for integrating species traits, phylogeny, and compositional effects [261, 262], and multivariate point process models for capturing spatial dependencies in species distributions [263]. SPIEC-EASI, which uses sparse inverse covariance estimation under compositional constraints [264]. FlashWeave, designed to detect both direct and indirect associations in cross-sectional and longitudinal data [265], and CoNet, an ensemble method integrating multiple correlation and dissimilarity metrics [266]. These approaches examine species dispersal and migration in microbiomes that also drive the evolutionary dynamics and acquire functional adaptations over time [267–269]. Additionally, host-mediated microbiome interaction models integrate host physiology, immune responses, and microbiome function, bridging the gap between microbial community behavior and host-driven evolution of microbial composition, ultimately influencing systemic health outcomes [65, 270, 271].

Table 2. Comparative overview of community-scale model types.

Model type	Lumped	Compartment-based	Nested
Core principle	Treats the entire community as a single metabolic unit	Includes multiple species as separate compartments in one model	Individual species models embedded in a higher-level community
Application	Initial screening of community metabolic capabilities and functional profiling.	Moderately detailed studies of multi-species interactions, focusing on cross-feeding and metabolite exchange between species.	Detailed studies of ecological and metabolic interactions, including dynamic ecosystem modeling and investigation of cross-feeding between species.
Objective function	Single biomass reaction representing the sum of all species' contributions. $v_{community\_biomass-}$	Single biomass reaction representing the weighted sum of species' biomass production $\sum w_i.biomass_{-i}$	It employs bi-level optimization: the lower level maximizes individual species growth ($v_{biomass,i}$) while the upper level optimizes overall community objectives $\sum w_i.biomass_{-i}$
Species representation	Species are not individually distinguished; the model predicts community-averaged behavior.	Each species is represented as a separate compartment.	Each species is modeled individually, then integrated to simulate community-level dynamics
Advantages	Simple to construct and computationally efficient. Aggregates community-wide metabolic potential.	Maintains species separation and allows metabolic exchange between species.	High-resolution modeling of individual and collective metabolism. Accounts for intra-/interspecies dynamics.
Limitations	Species-level interactions are not possible. Lacks taxonomic resolution.	Requires detailed genome annotations. Increases model complexity.	Requires detailed genome annotations. Computationally intensive. Data-demanding (omics, kinetic parameters)

Community-scale metabolic modeling and omics data integration

Microbial metabolic interactions are responsible for the fitness of interacting species, besides shaping the structure and function of microbial communities. This has implications in health ecology and microevolution [15, 42]. To gain valuable insights into the functional capabilities and mechanisms underlying various molecular interactions within or between microorganisms, genome-scale metabolic models (GEMs) have served as powerful tools [272, 273]. GEMs represent the complete set of metabolic reactions of an organism or community based on its genome, constructed by integrating genomic, biochemical, and physiological data and undergoing extensive refinement before being used as predictive tools [16, 274, 275]. GEMs of >7000 human microorganisms have been reconstructed that could be used to reconstruct community-based GEMs [276]. Integrating with metagenomics, metatranscriptomics, metaproteomics, and/or metabolomics data, community-scale models can guide the exploration of complex metabolic interactions, even if it doesn't capture all molecular or microbial details [238]. 'There are three main types of community-scale GEMs: the lumped model, the compartment-based model, and the nested community models' [272]. (Table 2, Fig. 3).

In the lumped-type models, also referred to as mixed-bag community modeling or gene-soup, reactions of each species of the community are pooled to create a single model. Such models contain a single copy of each metabolic reaction or metabolite, although that might be present in several species, and contain one combined biomass growth reaction made up of the growth reaction of all community members. These models are simple representations of microbial communities without species differentiation. It can be simulated by a regular flux balance analysis (FBA) approach, and provide insights about community growth, nutrient consumption, and chemical composition of the entire community. However, it cannot be used to examine species–species interactions within a community. The lumped-type metabolic network of the microbiome was used to identify gene and network differences linked to obesity and IBD [277]. Another study created a lumped metabolic model of the microbiome and showed differences in metabolite production capabilities between the two communities [84].

On the other hand, compartment-based GEMs create a single metabolic model of the community and treat each microbial species, with its metabolic network, as a compartment [241, 272, 278, 279]. This type of community model is very similar to the model of eukaryotic species with several internal compartments and genes, reactions, and metabolites of each species treated separately. In this type of model, the entire community interacts with the environment through a set of exchange reactions. Moreover, the transport reactions between species are allowed, which are crucial for studying metabolic interactions between species. Also, a single objective (mainly growth rate) is assumed for the entire community, in which the biomass components are the weighted biomass of individual species. The stoichiometry of community biomass can be set based on the abundance profile of species from metagenome experiments (Fig. 3). This approach is the most commonly used in modeling microbial communities. However, a common problem with this approach is that multiple flux combinations can achieve the same optimal result. Various approaches, such as the community FBA (cFBA) or MiCOM, address this issue. A study using genome-scale metabolic models identified metabolite exchange interactions between auxotrophic and prototrophic strains, revealing enhanced antimicrobial resilience in interacting partners [241]. Community-scale GEMs are used to analyze microbiome dynamics in IBD and gouty and rheumatoid arthritis patients [40, 279–282]. Another recent study combining computational models and metagenomics identified bacterial species exhibiting altered growth responses to metformin, highlighting metabolic pathways linked to the drug's hypoglycemic effects [141].

In contrast, nested community GEMs treat each species separately, constructing community-scale metabolic models hierarchically, where individual microbes are embedded within the higher-level microbiome. These models rely on bilevel optimization, which involves two interconnected optimization problems: one for the overall microbial community (e.g. maximizing community growth) and another for individual species (e.g. optimizing species-specific growth) (Fig. 3). This approach mimics real

microbial communities by allowing species to grow at different rates based on their objectives, preventing the unrealistic dominance of a few fast-growing species. Additionally, it enables the analysis of interspecies interactions. However, as community size increases, the complexity of the model also grows, making computational analysis more challenging. Using the CASINO (Community And Systems-level INteractive Optimization) toolbox, this approach was used in a diet-intervention study of obese and overweight individuals [283]. Integrating omics data can resolve complex metabolic interactions and refine existing genome-scale models [284, 285].

Some of the most common model reconstruction platforms are ModelSEED [286], Merlin [287], RAVEN [288], CarveMe [289], and gapseq [290], among others, and have been extensively used in modeling microbial species and communities. Quality control of GEMs is crucial, for that purpose, MEMOTE is a notable tool for improving the model accuracy [291]. ModelSEED generates fully automated models with a predefined biomass function, enabling phenotype simulations, while Merlin offers semi-automated reconstruction through a user-friendly graphical interface. RAVEN Toolbox 2.0 facilitates GEM reconstruction and simulations using reaction data from databases like KEGG and MetaCyc. CarveMe is another automated tool that efficiently constructs simulation-ready GEMs and is relatively faster than others. Similarly, gapseq creates automated GEMs using a curated reaction database and a novel gap-filling algorithm, improving model completeness and accuracy. Various computational platforms like the COBRA Toolbox [292], OptFlux, and KBase [293] are used to predict metabolic fluxes of microbial community species [274, 294]. Furthermore, the scope of GEMs will expand with continuous data curation, refined algorithms, and integration of multi-omics will enable a deeper understanding of microbiome function at the systems level. Lastly, GEMs are often integrated with various omics datasets to gain deeper insights into biological systems.

Challenges in multi-omics experiments and computational modeling approaches in microbiome research

Omics-based approaches have limitations and present their challenges. For instance, metagenomic analyses often fail to detect low-abundance microbes due to difficulties in sample isolation and underrepresentation in sequencing data, even though these taxa may play keystone roles in network stability and host interactions [295]. Sequence assembly is another challenge, as millions of short reads must be reconstructed into thousands of microbial genomes. Further, these assemblies require extensive computational resources for analysis and are often incomplete, resulting in metagenome-assembled genomes (MAGs). Long-read sequencing offers a potential solution to some of these issues. However, compared to short-read sequencing, it presents additional challenges, such as higher error rates, greater cost, lower throughput, higher DNA input requirements, and the need for specialized/dedicated bioinformatic tools. Moreover, there is no standardized pipeline for metagenomic data processing, and batch effects or confounding factors are difficult to eliminate. Another limitation of metagenomics is that, while it enables the identification of genes or gene families, assigning these genes to specific species requires additional computational processing such as assembly and binning, which are subject to limitations in accuracy, completeness, and taxonomic misclassifications. Metatranscriptomic and metaproteomic experiments introduce additional technical and analytical complexities. Extracting RNA or proteins from complex microbial communities is difficult due to differences in cell wall structures, and low-abundance transcripts or proteins may be lost during extraction. In metatranscriptomics, rRNA often dominates the dataset, masking mRNA signals and complicating downstream analyses. Furthermore, mapping sequence reads to reference genomes is limited by the availability of complete genome sequences. Metaproteomics faces similar obstacles, particularly in detecting low-abundance microbial proteins. Assigning peptides to specific microbial species remains a challenge, as identified peptides often lack species-specific markers. These issues make it difficult to accurately interpret metatranscriptomic and metaproteomic data, necessitating further methodological advancements and validation approaches [296].

Similarly, constructing and simulating mathematical models of microbial communities presents several challenges. Due to the complexity of microbial communities, most studies focus only on highly abundant species, often overlooking the potential influence of low-abundance microbes [297, 298]. However, these low-abundance microbes can play key roles in the community, such as helping with nutrient cycling, controlling harmful microbes, or supporting the immune system [299]. Additionally, uncultured microbes are often excluded from computational models due to limited data, and the vast diversity and complex interactions within microbial communities make accurate ecological or metabolic modeling challenging. In community-based modeling, networks are typically constructed using gene pool data or species abundance profiles. Simulations are then performed based on overall community growth or a combination of individual species and community growth [297]. However, the model prediction can achieve the same objective in multiple ways and often yields multiple possible solutions, making interpretation difficult. Furthermore, the experimental validation of model predictions is difficult due to the lack of direct measurements in microbial ecosystems [298, 300].

Integrative systems biology approaches for characterizing the microbiome and advancing future microbiome research

Integrative systems biology, which combines omics datasets with mathematical models, is essential for uncovering microbial community phenotypes and species interactions. For instance, whole-genome metagenomics provides gene pool profiles from different microbiomes, which can be used to construct context-specific community models by selectively retaining relevant reactions [208] (Fig. 3). This allows for more realistic predictions of metabolic fluxes, as demonstrated in numerous studies [301–313]. Additionally, species abundance profiles from amplicon-based or whole-genome microbiome studies can be mapped to ecological networks or metabolic models to determine biomass weight distributions and compare community dynamics across different environments [173, 207] (Fig. 3). Similarly, metatranscriptomics and metaproteomics data can be incorporated into community GEM models by adjusting reaction flux bounds based on gene or protein expression or reducing the network size to include only reactions associated with expressed genes [273, 314]. Despite limited experimental validation, this method can guide predictions toward more biologically relevant flux distributions, offering critical insights into community dynamics. Such integrative approaches have been widely applied to study metabolic interactions within and between microbial species, improving our understanding of microbiome functions and their impact on health and the environment [253, 315].

Engineering synthetic microbial communities that mimic specific conditions enables controlled modeling of interspecies interactions and microbial functions. Integrating synthetic microbial communities' models with omics datasets further enhances our understanding of microbiome dynamics. Validated predictions from these models can inform diagnostics and therapeutics through systems and synthetic biology [316–318]. Tools such as biosensors are engineered to respond to specific metabolites or environmental signals that enable real-time tracking of microbiome activity and host responses. Progress in molecular biology and genetics has further enabled the forward engineering and manipulation of the human-associated microbiota, offering new diagnostic and therapeutic strategies against diseases of microbial origin [319, 320]. Emerging technologies, including engineered organoids and high-throughput culturing, are revolutionizing gut microbiome research, significantly enhancing the efficiency and quality of studies [321]. For example, systems like the Intestinal Organoid Physoxic Coculture model enable physiologically relevant coculture with anaerobic microbes, supporting studies on epithelial responses, immune signaling, and the cultivation of previously unculturable gut bacteria [322, 323]. These advancements highlight a concerted effort to deepen our understanding of the microbiome's interactions with the host, paving the way for the development of personalized medical interventions and preventive strategies.

Quantum computing offers the potential to model complex microbial interactions by efficiently simulating high-dimensional, nonlinear systems, such as metabolic networks and community dynamics, that are computationally intensive for classical methods [324]. While classical approaches rely on step-by-step calculations, quantum algorithms can explore multiple states simultaneously, enabling faster solutions for problems like microbial network optimization or reaction flux prediction. Furthermore, quantum machine learning can enhance multi-omics analysis in microbial ecosystems by accelerating feature selection, improving dimensionality reduction, and capturing complex patterns across genomic, transcriptomic, and metabolomic layers [325, 326]. The shift from classical to quantum models will enable us to decrypt and interpret these interactions with unprecedented accuracy and speed, especially for phenomena such as antibiotic resistance and microbial signaling, suggesting that quantum models could one day capture not just the data structures of microbial ecosystems but aspects of their fundamental biophysics [327–329]. By embracing quantum machine learning, we can explore multi-omics data and reveal hidden patterns in microbial dynamics, paving the way for novel therapeutic interventions [330]. This interdisciplinary approach is not just a visionary concept but a necessary evolution in computational biology, with the promise of unraveling the complexity of microbial ecosystems in previously unimaginable ways [331, 332]. To advance the field of integrative computational and experimental microbiome research driven by an interactive cycle of computational modeling, it is necessary to let the field grow by enabling easy-to-use modeling frameworks to let nonexpert researchers perform their modeling. To stimulate these researchers, thereby expanding the scope of computational/hypothesis-driven research, recent advances in artificial intelligence (AI), such as AI coding assistance, may greatly stimulate research on the complex nature of microbiome research.

Conclusion

The diversity of the human microbiome is essential for health, performing a wide range of vital functions, and an abnormal change in the structure is known as microbiome dysbiosis and has been linked to various complex diseases. Several environmental and lifestyle factors contribute to microbiome dysbiosis, but targeted therapeutic interventions may help restore the microbiome to its normal state. In microbiome research, various traditional omics approaches, such as metagenomics, meta-transcriptomics, and meta-proteomics, are commonly used to study microbiome structure in disease conditions. Similarly, mathematical and computational approaches have been used to examine the mechanisms of diverse microbial interactions within the microbiome. Individual omics or computational models alone cannot fully capture microbiome complexity, whereas integrative systems biology combines both to provide a more comprehensive, mechanistic view of microbial dynamics. This framework enables tracing key interactions central to microbiome dynamics and identifies relevant biological markers. Emerging tools like quantum computing and artificial intelligence, particularly for modeling high-dimensional host microbiome systems, offer deeper insights into complex host–microbiome interactions, advancing omics analysis, improving computational models, and paving the way for novel diagnostics and therapeutics.

Key Points

- The human microbiome diversity plays an essential role in maintaining health.
- Microbiome dysbiosis is linked to numerous complex disorders.
- Omics and computational models offer insights into microbial function and interactions.
- Integrative systems biology utilizes omics data and computational models, provides a mechanistic understanding of microbial community dynamics, and identifies biologically relevant markers.
- Emerging technologies, such as quantum computing and artificial intelligence, may transform microbiome analysis.

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Author contributions

M.T.A. conceptualized the review idea and wrote the first draft with S.S.R. All authors contributed to writing.

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Data availability

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

References

1. Lee J-Y, Tsolis RM, Bäumler AJ. The microbiome and gut homeostasis. *Science* 2022;**377**:eabp9960. https://doi.org/10.1126/science.abp9960.
2. Lozupone CA, Stombaugh JI, Gordon JI. *et al.* Diversity, stability and resilience of the human gut microbiota. *Nature* 2012;**489**:220–30. https://doi.org/10.1038/nature11550.
3. Madi N, Cato ET, Sayeed A. *et al.* Phage predation, disease severity, and pathogen genetic diversity in cholera patients. *Science* 2024;**384**:292–2. https://doi.org/10.1126/science.adj3166.
4. Manor O, Dai CL, Kornilov SA. *et al.* Health and disease markers correlate with gut microbiome composition across thousands of people. *Nat Commun* 2020;**11**:5206. https://doi.org/10.1038/s41467-020-18871-1.
5. Haran JP, McCormick BA. Aging, frailty, and the microbiome—How dysbiosis influences human aging and disease. *Gastroenterology* 2021;**160**:507–23. https://doi.org/10.1053/j.gastro.2020.09.060.
6. Maier L, Goemans CV, Wirbel J. *et al.* Unravelling the collateral damage of antibiotics on gut bacteria. *Nature* 2021;**599**:120–4. https://doi.org/10.1038/s41586-021-03986-2.
7. Wilde J, Slack E, Foster KR. Host control of the microbiome: Mechanisms, evolution, and disease. *Science* 2024;**385**:eadi3338. https://doi.org/10.1126/science.adi3338.
8. Ross FC, Patangia D, Grimaud G. *et al.* The interplay between diet and the gut microbiome: Implications for health and disease. *Nat Rev Microbiol* 2024;**22**:671–86. https://doi.org/10.1038/s41579-024-01068-4.
9. De Filippis F, Valentino V, Sequino G. *et al.* Exposure to environmental pollutants selects for xenobiotic-degrading functions in the human gut microbiome. *Nat Commun* 2024;**15**:4482. https://doi.org/10.1038/s41467-024-48739-7.
10. Lippert K, Kedenko L, Antonielli L. *et al.* Gut microbiota dysbiosis associated with glucose metabolism disorders and the metabolic syndrome in older adults. *Benefic Microbes* 2017;**8**:545–56. https://doi.org/10.3920/BM2016.0184.
11. Keshet A, Segal E. Identification of gut microbiome features associated with host metabolic health in a large population-based cohort. *Nat Commun* 2024;**15**:9358. https://doi.org/10.1038/s41467-024-53832-y.
12. Yin Q, da Silva AC, Zorrilla F. *et al.* Ecological dynamics of Enterobacteriaceae in the human gut microbiome across global populations. *Nat Microbiol* 2025;**10**:541–53. https://doi.org/10.1038/s41564-024-01912-6.
13. Fogelson KA, Dorrestein PC, Zarrinpar A. *et al.* The gut microbial bile acid modulation and its relevance to digestive health and diseases. *Gastroenterology* 2023;**164**:1069–85. https://doi.org/10.1053/j.gastro.2023.02.022.
14. Schluter J, Peled JU, Taylor BP. *et al.* The gut microbiota is associated with immune cell dynamics in humans. *Nature* 2020;**588**:303–7. https://doi.org/10.1038/s41586-020-2971-8.
15. Li F, Chen Y, Gustafsson J. *et al.* Genome-scale metabolic models applied for human health and biopharmaceutical engineering. *Quant Biol* 2023;**11**:363–75. https://doi.org/10.1002/qub2.21.
16. Carter EL, Constantinidou C, Alam MT. Applications of genome-scale metabolic models to investigate microbial metabolic adaptations in response to genetic or environmental perturbations. *Brief Bioinform* 2024;**25**:bbad439.
17. Sender R, Fuchs S, Milo R. Are we really vastly outnumbered? Revisiting the ratio of bacterial to host cells in humans. *Cell* 2016;**164**:337–40. https://doi.org/10.1016/j.cell.2016.01.013.
18. Tierney BT, Yang Z, Luber JM. *et al.* The landscape of genetic content in the gut and oral human microbiome. *Cell Host Microbe* 2019;**26**:283–295.e8. https://doi.org/10.1016/j.chom.2019.07.008.
19. Eckburg PB, Bik EM, Bernstein CN. *et al.* Diversity of the human intestinal microbial Flora. *Science* 2005;**308**:1635–8. https://doi.org/10.1126/science.1110591.
20. Ma Z, Zuo T, Frey N. *et al.* A systematic framework for understanding the microbiome in human health and disease: From basic principles to clinical translation. *Signal Transduct Target Ther* 2024;**9**:1–36. https://doi.org/10.1038/s41392-024-01946-6.
21. Constantinides MG, Belkaid Y. Early-life imprinting of unconventional T cells and tissue homeostasis. *Science* 2021;**374**:eabf0095. https://doi.org/10.1126/science.abf0095.
22. Bogaert D, Beveren GJ, De VKEM. *et al.* Mother-to-infant microbiota transmission and infant microbiota development across multiple body sites. *Cell Host Microbe* 2023;**31**:447–460.e6. https://doi.org/10.1016/j.chom.2023.01.018.
23. Kennedy MS, Chang EB. The microbiome: Composition and locations. *Prog Mol Biol Transl Sci* 2020;**176**:1–42. https://doi.org/10.1016/bs.pmbts.2020.08.013.
24. Byrd AL, Belkaid Y, Segre JA. The human skin microbiome. *Nat Rev Microbiol* 2018;**16**:143–55. https://doi.org/10.1038/nrmicro.2017.157.
25. Mayhew D, Devos N, Lambert C. *et al.* Longitudinal profiling of the lung microbiome in the AERIS study demonstrates repeatability of bacterial and eosinophilic COPD exacerbations. *Thorax* 2018;**73**:422–30. https://doi.org/10.1136/thoraxjnl-2017-210408.
26. Li R, Li J, Zhou X. Lung microbiome: New insights into the pathogenesis of respiratory diseases. *Signal Transduct Target Ther* 2024;**9**:1–27. https://doi.org/10.1038/s41392-023-01668-1.
27. Kim YJ, Bunyavanich S. Microbial influencers: The airway microbiome's role in asthma. *J Clin Invest* 2025;**135**:e184316. https://doi.org/10.1172/JCI184316.
28. Wang Z, Singh R, Miller BE. *et al.* Sputum microbiome temporal variability and dysbiosis in chronic obstructive pulmonary disease exacerbations: An analysis of the COPDMAP study. *Thorax* 2018;**73**:331–8. https://doi.org/10.1136/thoraxjnl-2017-210741.
29. Bahetjan K, Yu-Xia LS, Lin S. *et al.* Analysis of the bronchoalveolar lavage fluid microbial flora in COPD patients at different lung function during acute exacerbation. *Sci Rep* 2025;**15**:13179. https://doi.org/10.1038/s41598-025-96746-5.
30. Lee CY, Dillard LR, Papin JA. *et al.* New perspectives into the vaginal microbiome with systems biology. *Trends Microbiol* 2023;**31**:356–68. https://doi.org/10.1016/j.tim.2022.09.011.
31. Liao J, Shenhav L, Urban JA. *et al.* Microdiversity of the vaginal microbiome is associated with preterm birth. *Nat Commun* 2023;**14**:4997. https://doi.org/10.1038/s41467-023-40719-7.
32. Belkaid Y, Harrison OJ. Homeostatic immunity and the microbiota. *Immunity* 2017;**46**:562–76. https://doi.org/10.1016/j.immuni.2017.04.008.
33. Li T-T, Chen X, Huo D. *et al.* Microbiota metabolism of intestinal amino acids impacts host nutrient homeostasis and physiology. *Cell Host Microbe* 2024;**32**:661–675.e10. https://doi.org/10.1016/j.chom.2024.04.004.
34. Byndloss MX, Olsan EE, Rivera-Chávez F. *et al.* Microbiota-activated PPAR-γ signaling inhibits dysbiotic Enterobacteriaceae expansion. *Science* 2017;**357**:570–5. https://doi.org/10.1126/science.aam9949.
35. Valles-Colomer M. The neuroactive potential of the human gut microbiota in quality of life and depression. *Nat Microbiol* 2019;**4**:623–32. https://doi.org/10.1038/s41564-018-0337-x.

36. Jacobson A, Yang D, Vella M. *et al.* The intestinal neuroimmune axis: Crosstalk between neurons, immune cells, and microbes. *Mucosal Immunol* 2021;**14**:555–65. https://doi.org/10.1038/s41385-020-00368-1.
37. Loh JS, Mak WQ, Tan LKS. *et al.* Microbiota–gut–brain axis and its therapeutic applications in neurodegenerative diseases. *Signal Transduct Target Ther* 2024;**9**:1–53. https://doi.org/10.1038/s41392-024-01743-1.
38. Tofani GSS, Leigh S-J, Gheorghe CE. *et al.* Gut microbiota regulates stress responsivity via the circadian system. *Cell Metab* 2025;**37**:138–153.e5. https://doi.org/10.1016/j.cmet.2024.10.003.
39. Mani A, Henn C, Couch C. *et al.* A brain microbiome in salmonids at homeostasis. *Sci Adv* 2024;**10**:eado0277. https://doi.org/10.1126/sciadv.ado0277.
40. O'Riordan KJ, Moloney GM, Keane L. *et al.* The gut microbiota-immune-brain axis: Therapeutic implications. *Cell Rep Med* 2025;**6**:101982. https://doi.org/10.1016/j.xcrm.2025.101982.
41. Alam MT, Amos GCA, Murphy ARJ. *et al.* Microbial imbalance in inflammatory bowel disease patients at different taxonomic levels. *Gut Pathog* 2020;**12**:1. https://doi.org/10.1186/s13099-019-0341-6.
42. Kost C, Patil KR, Friedman J. *et al.* Metabolic exchanges are ubiquitous in natural microbial communities. *Nat Microbiol* 2023;**8**:2244–52. https://doi.org/10.1038/s41564-023-01511-x.
43. Donohoe DR, Garge N, Zhang X. *et al.* The microbiome and butyrate regulate energy metabolism and autophagy in the mammalian colon. *Cell Metab* 2011;**13**:517–26. https://doi.org/10.1016/j.cmet.2011.02.018.
44. Chang PV, Hao L, Offermanns S. *et al.* The microbial metabolite butyrate regulates intestinal macrophage function via histone deacetylase inhibition. *Proc Natl Acad Sci* 2014;**111**:2247–52. https://doi.org/10.1073/pnas.1322269111.
45. Sanna S, van Zuydam NR, Mahajan A. *et al.* Causal relationships among the gut microbiome, short-chain fatty acids and metabolic diseases. *Nat Genet* 2019;**51**:600–5. https://doi.org/10.1038/s41588-019-0350-x.
46. Kropp C, Le Corf K, Relizani K. *et al.* The keystone commensal bacterium Christensenella minuta DSM 22607 displays anti-inflammatory properties both in vitro and in vivo. *Sci Rep* 2021;**11**:11494. https://doi.org/10.1038/s41598-021-90885-1.
47. Kim S, Seo S-U, Kweon M-N. Gut microbiota-derived metabolites tune host homeostasis fate. *Semin Immunopathol* 2024;**46**:2. https://doi.org/10.1007/s00281-024-01012-x.
48. Mukhopadhya I, Louis P. Gut microbiota-derived short-chain fatty acids and their role in human health and disease. *Nat Rev Microbiol* 2025;**1–17**. https://doi.org/10.1038/s41579-025-01183-w.
49. Gentile CL, Weir TL. The gut microbiota at the intersection of diet and human health. *Science* 2018;**362**:776–80. https://doi.org/10.1126/science.aau5812.
50. Kang WK, Florman JT, Araya A. *et al.* Vitamin B12 produced by gut bacteria modulates cholinergic signalling. *Nat Cell Biol* 2024;**26**:72–85. https://doi.org/10.1038/s41556-023-01299-2.
51. Grant ET, Parrish A, Boudaud M. *et al.* Dietary fibers boost gut microbiota-produced B vitamin pool and alter host immune landscape. *Microbiome* 2024;**12**:179. https://doi.org/10.1186/s40168-024-01898-7.
52. Ellis JL, Karl JP, Oliverio AM. *et al.* Dietary vitamin K is remodeled by gut microbiota and influences community composition. *Gut Microbes* 2021;**13**:1–16. https://doi.org/10.1080/19490976.2021.1887721.
53. Singh P, Rawat A, Alwakeel M. *et al.* The potential role of vitamin D supplementation as a gut microbiota modifier in healthy individuals. *Sci Rep* 2020;**10**:21641. https://doi.org/10.1038/s41598-020-77806-4.
54. Routy B, Le Chatelier E, Derosa L. *et al.* Gut microbiome influences efficacy of PD-1–based immunotherapy against epithelial tumors. *Science* 2018;**359**:91–7. https://doi.org/10.1126/science.aan3706.
55. Lynch JB, Hsiao EY. Microbiomes as sources of emergent host phenotypes. *Science* 2019;**365**:1405–9. https://doi.org/10.1126/science.aay0240.
56. Mallott EK, Amato KR. Host specificity of the gut microbiome. *Nat Rev Microbiol* 2021;**19**:639–53. https://doi.org/10.1038/s41579-021-00562-3.
57. Zhou X, Shen X, Johnson JS. *et al.* Longitudinal profiling of the microbiome at four body sites reveals core stability and individualized dynamics during health and disease. *Cell Host Microbe* 2024;**32**:506–526.e9. https://doi.org/10.1016/j.chom.2024.02.012.
58. Ziegler M, Seneca FO, Yum LK. *et al.* Bacterial community dynamics are linked to patterns of coral heat tolerance. *Nat Commun* 2017;**8**:14213. https://doi.org/10.1038/ncomms14213.
59. de Wit DF, Hanssen NMJ, Wortelboer K. *et al.* Evidence for the contribution of the gut microbiome to obesity and its reversal. *Sci Transl Med* 2023;**15**:eadg2773. https://doi.org/10.1126/scitranslmed.adg2773.
60. De Vos WM, Tilg H, Van Hul M. *et al.* Gut microbiome and health: Mechanistic insights. *Gut* 2022;**71**:1020–32. https://doi.org/10.1136/gutjnl-2021-326789.
61. Makki K, Deehan EC, Walter J. *et al.* The impact of dietary Fiber on gut microbiota in host health and disease. *Cell Host Microbe* 2018;**23**:705–15. https://doi.org/10.1016/j.chom.2018.05.012.
62. Deehan EC, Mocanu V, Madsen KL. Effects of dietary fibre on metabolic health and obesity. *Nat Rev Gastroenterol Hepatol* 2024;**21**:301–18.
63. Fackelmann G, Manghi P, Carlino N. *et al.* Gut microbiome signatures of vegan, vegetarian and omnivore diets and associated health outcomes across 21,561 individuals. *Nat Microbiol* 2025;**10**:41–52. https://doi.org/10.1038/s41564-024-01870-z.
64. Shuai M, Zhang G, Zeng F. *et al.* Human gut antibiotic Resistome and progression of diabetes. *Adv Sci* 2022;**9**:2104965. https://doi.org/10.1002/advs.202104965.
65. Singh S, Giron LB, Shaikh MW. *et al.* Distinct intestinal microbial signatures linked to accelerated systemic and intestinal biological aging. *Microbiome* 2024;**12**:31. https://doi.org/10.1186/s40168-024-01758-4.
66. Dong X, Pan P, Zheng D-W. *et al.* Bioinorganic hybrid bacteriophage for modulation of intestinal microbiota to remodel tumor-immune microenvironment against colorectal cancer. *Sci Adv* 2020;**6**:eaba1590. https://doi.org/10.1126/sciadv.aba1590.
67. Foster JA, Rinaman L, Cryan JF. Stress & the gut-brain axis: Regulation by the microbiome. *Neurobiol Stress* 2017;**7**:124–36. https://doi.org/10.1016/j.ynstr.2017.03.001.
68. Ke S, Wang X-W, Ratanatharathorn A. *et al.* Association of probable post-traumatic stress disorder with dietary pattern and gut microbiome in a cohort of women. *Nat Ment Health* 2023;**1**:900–13. https://doi.org/10.1038/s44220-023-00145-6.
69. Matenchuk BA, Mandhane PJ, Kozyrskyj AL. Sleep, circadian rhythm, and gut microbiota. *Sleep Med Rev* 2020;**53**:101340. https://doi.org/10.1016/j.smrv.2020.101340.

70. Maghini DG, Oduaran OH, Olubayo LAI. *et al.* Expanding the human gut microbiome atlas of Africa. *Nature* 2025;**638**:718–28.
71. Pasolli E, Asnicar F, Manara S. *et al.* Extensive unexplored human microbiome diversity revealed by over 150,000 genomes from metagenomes spanning age, geography, and lifestyle. *Cell* 2019;**176**:649–662.e20. https://doi.org/10.1016/j.cell.2019.01.001.
72. Gacesa R. Environmental factors shaping the gut microbiome in a Dutch population. *Nature* 2022;**604**:732–9. https://doi.org/10.1038/s41586-022-04567-7.
73. Feng Q, Liang S, Jia H. *et al.* Gut microbiome development along the colorectal adenoma–carcinoma sequence. *Nat Commun* 2015;**6**:6528. https://doi.org/10.1038/ncomms7528.
74. Cani PD, Amar J, Iglesias MA. *et al.* Metabolic endotoxemia initiates obesity and insulin resistance. *Diabetes* 2007;**56**:1761–72. https://doi.org/10.2337/db06-1491.
75. Fromentin S, Forslund SK, Chechi K. *et al.* Microbiome and metabolome features of the cardiometabolic disease spectrum. *Nat Med* 2022;**28**:303–14. https://doi.org/10.1038/s41591-022-01688-4.
76. Sampson TR, Debelius JW, Thron T. *et al.* Gut microbiota regulate motor deficits and neuroinflammation in a model of Parkinson's disease. *Cell* 2016;**167**:1469–1480.e12.
77. Shahin K, Soleimani-Delfan A, He Z. *et al.* Metagenomics revealed a correlation of gut phageome with autism spectrum disorder. *Gut Pathog* 2023;**15**:39. https://doi.org/10.1186/s13099-023-00561-0.
78. Loomba R, Seguritan V, Li W. *et al.* Gut microbiome-based metagenomic signature for non-invasive detection of advanced fibrosis in human nonalcoholic fatty liver disease. *Cell Metab* 2017;**25**:1054–1062.e5. https://doi.org/10.1016/j.cmet.2017.04.001.
79. Alpizar-Rodriguez D, Lesker TR, Gronow A. *et al.* Prevotella copri in individuals at risk for rheumatoid arthritis. *Ann Rheum Dis* 2019;**78**:590–3. https://doi.org/10.1136/annrheumdis-2018-214514.
80. Mekadim C, Skalnikova HK, Cizkova J. *et al.* Dysbiosis of skin microbiome and gut microbiome in melanoma progression. *BMC Microbiol* 2022;**22**:63. https://doi.org/10.1186/s12866-022-02458-5.
81. Sheflin AM, Whitney AK, Weir TL. Cancer-promoting effects of microbial dysbiosis. *Curr Oncol Rep* 2014;**16**:406. https://doi.org/10.1007/s11912-014-0406-0.
82. Ni J, Wu GD, Albenberg L. *et al.* Gut microbiota and IBD: Causation or correlation? *Nat Rev Gastroenterol Hepatol* 2017;**14**:573–84. https://doi.org/10.1038/nrgastro.2017.88.
83. Vestergaard MV, Allin KH, Eriksen C. *et al.* Gut microbiota signatures in inflammatory bowel disease. *United European Gastroenterol J* 2024;**12**:22–33. https://doi.org/10.1002/ueg2.12485.
84. Kumar M, Ji B, Babaei P. *et al.* Gut microbiota dysbiosis is associated with malnutrition and reduced plasma amino acid levels: Lessons from genome-scale metabolic modeling. *Metab Eng* 2018;**49**:128–42.
85. Yu T, Guo F, Yu Y. *et al.* Fusobacterium nucleatum promotes chemoresistance to colorectal cancer by modulating autophagy. *Cell* 2017;**170**:548–563.e16. https://doi.org/10.1016/j.cell.2017.07.008.
86. Sepich-Poore GD, Zitvogel L, Straussman R. *et al.* The microbiome and human cancer. *Science* 2021;**371**:eabc4552. https://doi.org/10.1126/science.abc4552.
87. Zepeda-Rivera M, Minot SS, Bouzek H. *et al.* A distinct fusobacterium nucleatum clade dominates the colorectal cancer niche. *Nature* 2024;**628**:424–32.
88. Berg G, Rybakova D, Fischer D. *et al.* Microbiome definition revisited: Old concepts and new challenges. *Microbiome* 2020;**8**:1–22.
89. Wu Y, Jiao N, Zhu R. *et al.* Identification of microbial markers across populations in early detection of colorectal cancer. *Nat Commun* 2021;**12**:3063. https://doi.org/10.1038/s41467-021-23265-y.
90. Coker OO, Liu C, Wu WKK. *et al.* Altered gut metabolites and microbiota interactions are implicated in colorectal carcinogenesis and can be non-invasive diagnostic biomarkers. *Microbiome* 2022;**10**:35. https://doi.org/10.1186/s40168-021-01208-5.
91. Iadsee N, Chuaypen N, Techawiwattanaboon T. *et al.* Identification of a novel gut microbiota signature associated with colorectal cancer in Thai population. *Sci Rep* 2023;**13**:6702. https://doi.org/10.1038/s41598-023-33794-9.
92. Anhê FF, Jensen BAH, Varin TV. *et al.* Type 2 diabetes influences bacterial tissue compartmentalisation in human obesity. *Nat Metab* 2020;**2**:233–42. https://doi.org/10.1038/s42255-020-0178-9.
93. Mardinoglu A. Plasma mannose levels are associated with incident type 2 diabetes and cardiovascular disease. *Cell Metab* 2017;**26**:281–3. https://doi.org/10.1016/j.cmet.2017.07.006.
94. Zhou W, Cheng Y, Zhu P. *et al.* Implication of gut microbiota in cardiovascular diseases. *Oxidative Med Cell Longev* 2020;**2020**:e5394096. https://doi.org/10.1155/2020/5394096.
95. Deng K, Xu J, Shen L. *et al.* Comparison of fecal and blood metabolome reveals inconsistent associations of the gut microbiota with cardiometabolic diseases. *Nat Commun* 2023;**14**:1–12. https://doi.org/10.1038/s41467-023-36256-y.
96. Ferrell M, Wang Z, Anderson JT. *et al.* A terminal metabolite of niacin promotes vascular inflammation and contributes to cardiovascular disease risk. *Nat Med* 2024;**30**:424–34. https://doi.org/10.1038/s41591-023-02793-8.
97. Meckel KR, Simpson SS, Godino A. *et al.* Microbial short-chain fatty acids regulate drug seeking and transcriptional control in a model of cocaine seeking. *Neuropsychopharmacology* 2024;**49**:386–95. https://doi.org/10.1038/s41386-023-01661-w.
98. Ahrens AP, Hyötyläinen T, Petrone JR. *et al.* Infant microbes and metabolites point to childhood neurodevelopmental disorders. *Cell* 2024;**187**:1853–1873.e15. https://doi.org/10.1016/j.cell.2024.02.035.
99. Braniste V, Al-Asmakh M, Kowal C. *et al.* The gut microbiota influences blood-brain barrier permeability in mice. *Sci Transl Med* 2014;**6**:158–263 158. https://doi.org/10.1126/scitranslmed.3009759.
100. Sharon G, Cruz NJ, Kang D-W. *et al.* Human gut microbiota from autism Spectrum disorder promote Behavioral symptoms in mice. *Cell* 2019;**177**:1600–1618.e17. https://doi.org/10.1016/j.cell.2019.05.004.
101. Yu W, Xiao Y, Jayaraman A. *et al.* Microbial metabolites tune amygdala neuronal hyperexcitability and anxiety-linked behaviors. *EMBO Mol Med* 2025;**17**:249–64. https://doi.org/10.1038/s44321-024-00179-y.
102. Hill-Burns EM, Debelius JW, Morton JT. *et al.* Parkinson's disease and Parkinson's disease medications have distinct signatures of the gut microbiome. *Mov Disord* 2017;**32**:739–49. https://doi.org/10.1002/mds.26942.
103. Pellegrini C, D'Antongiovanni V, Miraglia F. *et al.* Enteric α-synuclein impairs intestinal epithelial barrier through caspase-1-inflammasome signaling in Parkinson's disease before brain pathology. *Npj Park Dis* 2022;**8**:9. https://doi.org/10.1038/s41531-021-00263-x.

104. Zhao Z-H, Xin F-Z, Xue Y. *et al.* Indole-3-propionic acid inhibits gut dysbiosis and endotoxin leakage to attenuate steatohepatitis in rats. *Exp Mol Med* 2019;**51**:1–14. https://doi.org/10.1038/s12276-019-0304-5.
105. Hoyles L, Pontifex MG, Rodriguez-Ramiro I. *et al.* Regulation of blood–brain barrier integrity by microbiome-associated methylamines and cognition by trimethylamine N-oxide. *Microbiome* 2021;**9**:235. https://doi.org/10.1186/s40168-021-01181-z.
106. Xie J, Bruggeman A, De Nolf C. *et al.* Gut microbiota regulates blood-cerebrospinal fluid barrier function and aβ pathology. *EMBO J* 2023;**42**:e111515. https://doi.org/10.15252/embj.2022111515.
107. Liu C, Makrinioti H, Saglani S. *et al.* Microbial dysbiosis and childhood asthma development: Integrated role of the airway and gut microbiome, environmental exposures, and host metabolic and immune response. *Front Immunol* 2022;**13**:1028209. https://doi.org/10.3389/fimmu.2022.1028209.
108. Zhou Y, Jackson D, Bacharier LB. *et al.* The upper-airway microbiota and loss of asthma control among asthmatic children. *Nat Commun* 2019;**10**:5714. https://doi.org/10.1038/s41467-019-13698-x.
109. Barcik W, Boutin RCT, Sokolowska M. *et al.* The role of lung and gut microbiota in the pathology of asthma. *Immunity* 2020;**52**:241–55. https://doi.org/10.1016/j.immuni.2020.01.007.
110. Marsland BJ, Trompette A, Gollwitzer ES. The gut–lung Axis in respiratory disease. *Ann Am Thorac Soc* 2015;**12**:S150–6. https://doi.org/10.1513/AnnalsATS.201503-133AW.
111. Thompson KN, Bonham KS, Ilott NE. *et al.* Alterations in the gut microbiome implicate key taxa and metabolic pathways across inflammatory arthritis phenotypes. *Sci Transl Med* 2023;**15**:eabn4722. https://doi.org/10.1126/scitranslmed.abn4722.
112. Fyhrquist N, Muirhead G, Prast-Nielsen S. *et al.* Microbe-host interplay in atopic dermatitis and psoriasis. *Nat Commun* 2019;**10**:4703. https://doi.org/10.1038/s41467-019-12253-y.
113. Harris-Tryon TA, Grice EA. Microbiota and maintenance of skin barrier function. *Science* 2022;**376**:940–5. https://doi.org/10.1126/science.abo0693.
114. Sonnert ND, Rosen CE, Ghazi AR. *et al.* A host–microbiota interactome reveals extensive transkingdom connectivity. *Nature* 2024;**628**:171–9. https://doi.org/10.1038/s41586-024-07162-0.
115. Yang Y, Olah P, Radai Z. *et al.* Exploratory multi-omics analysis reveals host-microbe interactions associated with disease severity in psoriatic skin. *EBioMedicine* 2024;**105**:105222. https://doi.org/10.1016/j.ebiom.2024.105222.
116. Fassarella M, Blaak EE, Penders J. *et al.* Gut microbiome stability and resilience: Elucidating the response to perturbations in order to modulate gut health. *Gut* 2021;**70**:595–605. https://doi.org/10.1136/gutjnl-2020-321747.
117. El-Salhy M, Hatlebakk JG, Gilja OH. *et al.* Efficacy of faecal microbiota transplantation for patients with irritable bowel syndrome in a randomised, double-blind, placebo-controlled study. *Gut* 2020;**69**:859–67. https://doi.org/10.1136/gutjnl-2019-319630.
118. Spencer CN, McQuade JL, Gopalakrishnan V. *et al.* Dietary fiber and probiotics influence the gut microbiome and melanoma immunotherapy response. *Science* 2021;**374**:1632–40. https://doi.org/10.1126/science.aaz7015.
119. Medawar E, Beyer F, Thieleking R. *et al.* Prebiotic diet changes neural correlates of food decision-making in overweight adults: A randomised controlled within-subject cross-over trial. *Gut* 2024;**73**:298–310. https://doi.org/10.1136/gutjnl-2023-330365.
120. Wu H, Esteve E, Tremaroli V. *et al.* Metformin alters the gut microbiome of individuals with treatment-naive type 2 diabetes, contributing to the therapeutic effects of the drug. *Nat Med* 2017;**23**:850–8. https://doi.org/10.1038/nm.4345.
121. Strathdee SA, Hatfull GF, Mutalik VK. *et al.* Phage therapy: From biological mechanisms to future directions. *Cell* 2023;**186**:17–31. https://doi.org/10.1016/j.cell.2022.11.017.
122. Baldanzi G, Sayols-Baixeras S, Ekblom-Bak E. *et al.* Accelerometer-based physical activity is associated with the gut microbiota in 8416 individuals in SCAPIS. *EBioMedicine* 2024;**100**:104989.
123. van Lier YF, Davids M, Haverkate NJE. *et al.* Donor fecal microbiota transplantation ameliorates intestinal graft-versus-host disease in allogeneic hematopoietic cell transplant recipients. *Sci Transl Med* 2020;**12**:eaaz8926. https://doi.org/10.1126/scitranslmed.aaz8926.
124. Woodworth MH, Conrad RE, Haldopoulos M. *et al.* Fecal microbiota transplantation promotes reduction of antimicrobial resistance by strain replacement. *Sci Transl Med* 2023;**15**:eabo2750.
125. Kim J-H, Kim K, Kim W. Gut microbiota restoration through fecal microbiota transplantation: A new atopic dermatitis therapy. *Exp Mol Med* 2021;**53**:907–16. https://doi.org/10.1038/s12276-021-00627-6.
126. Bruggeman A, Vandendriessche C, Hamerlinck H. *et al.* Safety and efficacy of faecal microbiota transplantation in patients with mild to moderate Parkinson's disease (GUT-PARFECT): A double-blind, placebo-controlled, randomised, phase 2 trial. *eClinicalMedicine* 2024;**71**:102563. https://doi.org/10.1016/j.eclinm.2024.102563.
127. DeFilipp Z, Bloom PP, Soto MT. *et al.* Drug-resistant E. Coli Bacteremia transmitted by Fecal microbiota transplant. *N Engl J Med* 2019;**381**:2043–50. https://doi.org/10.1056/NEJMoa1910437.
128. Hanssen NMJ, De VWM, Nieuwdorp M. Fecal microbiota transplantation in human metabolic diseases: From a murky past to a bright future? *Cell Metab* 2021;**33**:1098–110. https://doi.org/10.1016/j.cmet.2021.05.005.
129. Quaranta G, Ianiro G, De Maio F. *et al.* "Bacterial consortium": A potential evolution of fecal microbiota transplantation for the treatment of Clostridioides difficile infection. *Biomed Res Int* 2022;**2022**:5787373. https://doi.org/10.1155/2022/5787373.
130. Furuichi M, Kawaguchi T, Pust M-M. *et al.* Commensal consortia decolonize Enterobacteriaceae via ecological control. *Nature* 2024;**633**:878–86. https://doi.org/10.1038/s41586-024-07960-6.
131. Bénard MV, De BCMA, Fenneman AC. *et al.* Challenges and costs of donor screening for fecal microbiota transplantations. *PLoS One* 2022;**17**:e0276323. https://doi.org/10.1371/journal.pone.0276323.
132. Khanna S, Assi M, Lee C. *et al.* Efficacy and Safety of RBX2660 in PUNCH CD3, a phase III, randomized, double-blind, placebo-controlled trial with a Bayesian primary analysis for the prevention of recurrent Clostridioides difficile infection. *Drugs* 2022;**82**:1527–38. https://doi.org/10.1007/s40265-022-01797-x.
133. Feuerstadt P, Louie TJ, Lashner B. *et al.* SER-109, an oral microbiome therapy for recurrent Clostridioides difficile infection. *N Engl J Med* 2022;**386**:220–9. https://doi.org/10.1056/NEJMoa2106516.
134. Kuziel GA, Lozano GL, Simian C. *et al.* Functional diversification of dietary plant small molecules by the gut microbiome. *Cell* 2025;**188**:1967–1983.e22. https://doi.org/10.1016/j.cell.2025.01.045.

135. Manghi P, Bhosle A, Wang K. *et al.* Coffee consumption is associated with intestinal Lawsonibacter asaccharolyticus abundance and prevalence across multiple cohorts. *Nat Microbiol* 2024;**9**:3120–34. https://doi.org/10.1038/s41564-024-01858-9.
136. Mohr AE, Sweazea KL, Bowes DA. *et al.* Gut microbiome remodeling and metabolomic profile improves in response to protein pacing with intermittent fasting versus continuous caloric restriction. *Nat Commun* 2024;**15**:4155.
137. Li X, Yang J, Zhou X. *et al.* Ketogenic diet-induced bile acids protect against obesity through reduced calorie absorption. *Nat Metab* 2024;**6**:1397–414. https://doi.org/10.1038/s42255-024-01072-1.
138. Bruce-Keller AJ, Richard AJ, Fernandez-Kim S-O. *et al.* Fenugreek counters the effects of high fat diet on gut microbiota in mice: Links to metabolic benefit. *Sci Rep* 2020;**10**:1245.
139. Moraïs S, Winkler S, Zorea A. *et al.* Cryptic diversity of cellulose-degrading gut bacteria in industrialized humans. *Science* 2024;**383**:eadj9223. https://doi.org/10.1126/science.adj9223.
140. Xie N, Zhang L, Gao W. *et al.* NAD+ metabolism: Pathophysiologic mechanisms and therapeutic potential. *Signal Transduct Target Ther* 2020;**5**:1–37. https://doi.org/10.1038/s41392-020-00311-7.
141. Ezzamouri B, Rosario D, Bidkhori G. *et al.* Metabolic modelling of the human gut microbiome in type 2 diabetes patients in response to metformin treatment. *Npj Syst Biol Appl* 2023;**9**:1–8. https://doi.org/10.1038/s41540-022-00261-6.
142. Crudele L, Gadaleta RM, Cariello M. *et al.* Gut microbiota in the pathogenesis and therapeutic approaches of diabetes. *EBioMedicine* 2023;**97**:104821.
143. Yan M, Man S, Sun B. *et al.* Gut liver brain axis in diseases: The implications for therapeutic interventions. *Signal Transduct Target Ther* 2023;**8**:1–26. https://doi.org/10.1038/s41392-023-01673-4.
144. Liang L, Su X, Guan Y. *et al.* Correlation between intestinal flora and GLP-1 receptor agonist dulaglutide in type 2 diabetes mellitus treatment—A preliminary longitudinal study. *iScience* 2024;**27**:109784. https://doi.org/10.1016/j.isci.2024.109784.
145. Dedrick RM, Smith BE, Cristinziano M. *et al.* Phage therapy of mycobacterium infections: Compassionate use of Phages in 20 patients with drug-resistant mycobacterial disease. *Clin Infect Dis* 2023;**76**:103–12. https://doi.org/10.1093/cid/ciac453.
146. Würstle S, Lee A, Kortright KE. *et al.* Optimized preparation pipeline for emergency phage therapy against Pseudomonas aeruginosa at Yale University. *Sci Rep* 2024;**14**:2657. https://doi.org/10.1038/s41598-024-52192-3.
147. Kim MK, Chen Q, Echterhof A. *et al.* A blueprint for broadly effective bacteriophage-antibiotic cocktails against bacterial infections. *Nat Commun* 2024;**15**:9987. https://doi.org/10.1038/s41467-024-53994-9.
148. Dubberke ER, Lee CH, Orenstein R. *et al.* Results from a randomized, placebo-controlled clinical trial of a RBX2660—A microbiota-based drug for the prevention of recurrent Clostridium difficile infection. *Clin Infect Dis* 2018;**67**:1198–204. https://doi.org/10.1093/cid/ciy259.
149. Abedon ST. Use of phage therapy to treat long-standing, persistent, or chronic bacterial infections. *Adv Drug Deliv Rev* 2019;**145**:18–39. https://doi.org/10.1016/j.addr.2018.06.018.
150. Advocating for phage therapy. *Nat Microbiol* 2024;**9**:1397–8. https://doi.org/10.1038/s41564-024-01733-7.
151. Deza DC, Alcedo J, Lafuente M. *et al. PROBIOTICS PRESCRIBED WITH HELICOBACTER PYLORI ERADICATION THERAPY IN EUROPE: USAGE PATTERN, EFFECTIVENESS, AND SAFETY: Results from the European Registry on Helicobacter Pylori Management (Hp-EuReg)*. Gastroenterol: Am. J, 2025.
152. Luo S-C, Wei S-M, Luo X-T. *et al.* How probiotics, prebiotics, synbiotics, and postbiotics prevent dental caries: An oral microbiota perspective. *Npj Biofilms Microbiomes* 2024;**10**:1–15.
153. Szajewska H, Scott KP, de Meij T. *et al.* Antibiotic-perturbed microbiota and the role of probiotics. *Nat Rev Gastroenterol Hepatol* 2024;**22**:155–72.
154. Scott BM, Gutiérrez-Vázquez C, Sanmarco LM. *et al.* Self-tunable engineered yeast probiotics for the treatment of inflammatory bowel disease. *Nat Med* 2021;**27**:1212–22. https://doi.org/10.1038/s41591-021-01390-x.
155. Praveschotinunt P, Duraj-Thatte AM, Gelfat I. *et al.* Engineered *E. coli* Nissle 1917 for the delivery of matrix-tethered therapeutic domains to the gut. *Nat Commun* 2019;**10**:5580. https://doi.org/10.1038/s41467-019-13336-6.
156. Liu J, Wang Y, Heelan WJ. *et al.* Mucoadhesive probiotic backpacks with ROS nanoscavengers enhance the bacteriotherapy for inflammatory bowel diseases. *Sci Adv* 2022;**8**:eabp8798.
157. Yang Y, Pan M, Xia X. *et al.* Effect of dietary probiotics intake on cancer mortality: A cohort study of NHANES 1999–2018. *Sci Rep* 2025;**15**:959. https://doi.org/10.1038/s41598-024-83722-8.
158. Prajapati SK, Wang S, Mishra SP. *et al.* Protection of Alzheimer's disease progression by a human-origin probiotics cocktail. *Sci Rep* 2025;**15**:1589. https://doi.org/10.1038/s41598-024-84780-8.
159. Ji J, Jin W, Liu S-J. *et al.* Probiotics, prebiotics, and postbiotics in health and disease. *MedComm* 2023;**4**:e420. https://doi.org/10.1002/mco2.420.
160. Yip AYG, King OG, Omelchenko O. *et al.* Antibiotics promote intestinal growth of carbapenem-resistant Enterobacteriaceae by enriching nutrients and depleting microbial metabolites. *Nat Commun* 2023;**14**:5094. https://doi.org/10.1038/s41467-023-40872-z.
161. Lindell AE, Zimmermann-Kogadeeva M, Patil KR. Multimodal interactions of drugs, natural compounds and pollutants with the gut microbiota. *Nat Rev Microbiol* 2022;**20**:431–43. https://doi.org/10.1038/s41579-022-00681-5.
162. Ricaurte D, Huang Y, Sheth RU. *et al.* High-throughput transcriptomics of 409 bacteria–drug pairs reveals drivers of gut microbiota perturbation. *Nat Microbiol* 2024;**9**:561–75. https://doi.org/10.1038/s41564-023-01581-x.
163. Yang X, Hang HC. Chemical genetic approaches to dissect microbiota mechanisms in health and disease. *Science* 2024;**386**:743–3. https://doi.org/10.1126/science.ado8548.
164. Zitvogel L, Ma Y, Raoult D. *et al.* The microbiome in cancer immunotherapy: Diagnostic tools and therapeutic strategies. *Science* 2018;**359**:1366–70. https://doi.org/10.1126/science.aar6918.
165. Wu H, Tremaroli V, Bäckhed F. Linking microbiota to human diseases: A systems biology perspective. *Trends Endocrinol Metab* 2015;**26**:758–70. https://doi.org/10.1016/j.tem.2015.09.011.
166. Li, G-. H. System-level metabolic modeling facilitates unveiling metabolic signature in exceptional longevity. *Aging Cell* 2022;**21**:e13595.
167. Kim N, Ma J, Kim W. *et al.* Genome-resolved metagenomics: A game changer for microbiome medicine. *Exp Mol Med* 2024;**56**:1501–12. https://doi.org/10.1038/s12276-024-01262-7.
168. Heintz-Buschart A, Wilmes P. Human gut microbiome: Function matters. *Trends Microbiol* 2018;**26**:563–74. https://doi.org/10.1016/j.tim.2017.11.002.
169. Knight R, Vrbanac A, Taylor BC. *et al.* Best practices for analysing microbiomes. *Nat Rev Microbiol* 2018;**16**:410–22. https://doi.org/10.1038/s41579-018-0029-9.
170. Zhang X, Li L, Butcher J. *et al.* Advancing functional and translational microbiome research using meta-omics approaches. *Microbiome* 2019;**7**:1–12.

171. Bahram M, Hildebrand F, Forslund SK. *et al.* Structure and function of the global topsoil microbiome. *Nature* 2018;**560**:233–7. https://doi.org/10.1038/s41586-018-0386-6.
172. Gupta S, Mortensen MS, Schjørring S. *et al.* Amplicon sequencing provides more accurate microbiome information in healthy children compared to culturing. *Commun Biol* 2019;**2**:1–7. https://doi.org/10.1038/s42003-019-0540-1.
173. Nearing JT, Douglas GM, Hayes MG. *et al.* Microbiome differential abundance methods produce different results across 38 datasets. *Nat Commun* 2022;**13**:342. https://doi.org/10.1038/s41467-022-28034-z.
174. Maljkovic Berry I, Melendrez MC, Bishop-Lilly KA. *et al.* Next generation sequencing and bioinformatics methodologies for infectious disease research and public health: Approaches, applications, and considerations for development of laboratory capacity. *J Infect Dis* 2020;**221**:S292–307. https://doi.org/10.1093/infdis/jiz286.
175. Louca S, Doebeli M, Parfrey LW. Correcting for 16S rRNA gene copy numbers in microbiome surveys remains an unsolved problem. *Microbiome* 2018;**6**:41. https://doi.org/10.1186/s40168-018-0420-9.
176. Gao Y, Wu M. Accounting for 16S rRNA copy number prediction uncertainty and its implications in bacterial diversity analyses. *ISME Commun* 2023;**3**:59. https://doi.org/10.1038/s43705-023-00266-0.
177. Liu S, Rodriguez JS, Munteanu V. *et al.* Analysis of metagenomic data. *Nat Rev Methods Primer* 2025;**5**:1–28. https://doi.org/10.1038/s43586-024-00376-6.
178. Lloréns-Rico V, Simcock JA, Huys GRB. *et al.* Single-cell approaches in human microbiome research. *Cell* 2022;**185**:2725–38. https://doi.org/10.1016/j.cell.2022.06.040.
179. Chijiiwa R, Hosokawa M, Kogawa M. *et al.* Single-cell genomics of uncultured bacteria reveals dietary fiber responders in the mouse gut microbiota. *Microbiome* 2020;**8**:5. https://doi.org/10.1186/s40168-019-0779-2.
180. Whitaker WR, Shepherd ES, Sonnenburg JL. Tunable expression tools enable single-cell strain distinction in the gut microbiome. *Cell* 2017;**169**:538–546.e12. https://doi.org/10.1016/j.cell.2017.03.041.
181. Sankaran K, Kodikara S, Li JJ. *et al.* Semisynthetic simulation for microbiome data analysis. *Brief Bioinform* 2025;**26**:bbaf051. https://doi.org/10.1093/bib/bbaf051.
182. Jia M, Zhu S, Xue M-Y. *et al.* Single-cell transcriptomics across 2,534 microbial species reveals functional heterogeneity in the rumen microbiome. *Nat Microbiol* 2024;**9**:1884–98. https://doi.org/10.1038/s41564-024-01723-9.
183. Zhu X, Huang X, Hu M. *et al.* A specific enterotype derived from gut microbiome of older individuals enables favorable responses to immune checkpoint blockade therapy. *Cell Host Microbe* 2024;**32**:489–505.e5. https://doi.org/10.1016/j.chom.2024.03.002.
184. Long S, Yang Y, Shen C. *et al.* Metaproteomics characterizes human gut microbiome function in colorectal cancer. *Npj Biofilms Microbiomes* 2020;**6**:1–10. https://doi.org/10.1038/s41522-020-0123-4.
185. Li L, Wang T, Ning Z. *et al.* Revealing proteome-level functional redundancy in the human gut microbiome using ultra-deep metaproteomics. *Nat Commun* 2023;**14**:3428. https://doi.org/10.1038/s41467-023-39149-2.
186. Tanca A, Palomba A, Fiorito G. *et al.* Metaproteomic portrait of the healthy human gut microbiota. *Npj Biofilms Microbiomes* 2024;**10**:1–12. https://doi.org/10.1038/s41522-024-00526-4.
187. Bauermeister A, Mannochio-Russo H, Costa-Lotufo LV. *et al.* Mass spectrometry-based metabolomics in microbiome investigations. *Nat Rev Microbiol* 2022;**20**:143–60. https://doi.org/10.1038/s41579-021-00621-9.
188. Lin H, Peddada SD. Analysis of compositions of microbiomes with bias correction. *Nat Commun* 2020;**11**:3514. https://doi.org/10.1038/s41467-020-17041-7.
189. Pinto Y, Bhatt AS. Sequencing-based analysis of microbiomes. *Nat Rev Genet* 2024;**25**:829–45. https://doi.org/10.1038/s41576-024-00746-6.
190. Herrera G, Castañeda S, Arboleda JC. *et al.* Metagenome-assembled genomes (MAGs) suggest an acetate-driven protective role in gut microbiota disrupted by Clostridioides difficile. *Microbiol Res* 2024;**285**:127739. https://doi.org/10.1016/j.micres.2024.127739.
191. Gloor GB, Macklaim JM, Pawlowsky-Glahn V. *et al.* Microbiome datasets are compositional: And this is not optional. *Front Microbiol* 2017;**8**:2224. https://doi.org/10.3389/fmicb.2017.02224.
192. Ha MJ, Kim J, Galloway-Peña J. *et al.* Compositional zero-inflated network estimation for microbiome data. *BMC Bioinformatics* 2020;**21**:581. https://doi.org/10.1186/s12859-020-03911-w.
193. Peterson CB, Saha S, Do K-A. Analysis of microbiome data. *Annu Rev Stat Its Appl* 2024;**11**:483–504. https://doi.org/10.1146/annurev-statistics-040522-120734.
194. Rausch P, Rühlemann M, Hermes BM. *et al.* Comparative analysis of amplicon and metagenomic sequencing methods reveals key features in the evolution of animal metaorganisms. *Microbiome* 2019;**7**:133. https://doi.org/10.1186/s40168-019-0743-1.
195. Bolyen E, Rideout JR, Dillon MR. *et al.* Reproducible, interactive, scalable and extensible microbiome data science using QIIME 2. *Nat Biotechnol* 2019;**37**:852–7. https://doi.org/10.1038/s41587-019-0209-9.
196. Callahan BJ, McMurdie PJ, Rosen MJ. *et al.* DADA2: High-resolution sample inference from Illumina amplicon data. *Nat Methods* 2016;**13**:581–3. https://doi.org/10.1038/nmeth.3869.
197. Schloss PD, Westcott SL, Ryabin T. *et al.* Introducing mothur: Open-source, platform-independent, community-supported software for describing and comparing microbial communities. *Appl Environ Microbiol* 2009;**75**:7537–41. https://doi.org/10.1128/AEM.01541-09.
198. Li D, Liu C-M, Luo R. *et al.* MEGAHIT: An ultra-fast single-node solution for large and complex metagenomics assembly via succinct *de Bruijn* graph. *Bioinformatics* 2015;**31**:1674–6. https://doi.org/10.1093/bioinformatics/btv033.
199. Nurk S, Meleshko D, Korobeynikov A. *et al.* metaSPAdes: A new versatile metagenomic assembler. *Genome Res* 2017;**27**:824–34. https://doi.org/10.1101/gr.213959.116.
200. Kang DD, Li F, Kirton E. *et al.* MetaBAT 2: An adaptive binning algorithm for robust and efficient genome reconstruction from metagenome assemblies. *PeerJ* 2019;**7**:e7359. https://doi.org/10.7717/peerj.7359.
201. Parks DH, Imelfort M, Skennerton CT. *et al.* CheckM: Assessing the quality of microbial genomes recovered from isolates, single cells, and metagenomes. *Genome Res* 2015;**25**:1043–55. https://doi.org/10.1101/gr.186072.114.
202. Mikheenko A, Saveliev V, Gurevich A. MetaQUAST: Evaluation of metagenome assemblies. *Bioinformatics* 2016;**32**:1088–90. https://doi.org/10.1093/bioinformatics/btv697.
203. Shaffer M, Borton MA, McGivern BB. *et al.* DRAM for distilling microbial metabolism to automate the curation of microbiome function. *Nucleic Acids Res* 2020;**48**:8883–900. https://doi.org/10.1093/nar/gkaa621.

204. Kolmogorov M, Bickhart DM, Behsaz B. *et al.* metaFlye: Scalable long-read metagenome assembly using repeat graphs. *Nat Methods* 2020;**17**:1103–10. https://doi.org/10.1038/s41592-020-00971-x.
205. Kim CY, Ma J, Lee I. HiFi metagenomic sequencing enables assembly of accurate and complete genomes from human gut microbiota. *Nat Commun* 2022;**13**:6367. https://doi.org/10.1038/s41467-022-34149-0.
206. Wood DE, Lu J, Langmead B. Improved metagenomic analysis with kraken 2. *Genome Biol* 2019;**20**:257. https://doi.org/10.1186/s13059-019-1891-0.
207. Truong DT, Franzosa EA, Tickle TL. *et al.* MetaPhlAn2 for enhanced metagenomic taxonomic profiling. *Nat Methods* 2016;**12**:902–3. https://doi.org/10.1038/nmeth.3589.
208. Beghini F, McIver LJ, Blanco-Míguez A. *et al.* Integrating taxonomic, functional, and strain-level profiling of diverse microbial communities with bioBakery 3. *elife* 2021;**10**:e65088. https://doi.org/10.7554/eLife.65088.
209. Patro R, Duggal G, Love MI. *et al.* Salmon provides fast and bias-aware quantification of transcript expression. *Nat Methods* 2017;**14**:417–9. https://doi.org/10.1038/nmeth.4197.
210. Bray NL, Pimentel H, Melsted P. *et al.* Near-optimal probabilistic RNA-seq quantification. *Nat Biotechnol* 2016;**34**:525–7. https://doi.org/10.1038/nbt.3519.
211. Love MI, Huber W, Anders S. Moderated estimation of fold change and dispersion for RNA-seq data with DESeq2. *Genome Biol* 2014;**15**:550. https://doi.org/10.1186/s13059-014-0550-8.
212. Robinson MD, McCarthy DJ, Smyth GK. edgeR: A Bioconductor package for differential expression analysis of digital gene expression data. *Bioinformatics* 2010;**26**:139–40. https://doi.org/10.1093/bioinformatics/btp616.
213. Cox J, Mann M. MaxQuant enables high peptide identification rates, individualized ppb-range mass accuracies and proteome-wide protein quantification. *Nat Biotechnol* 2008;**26**:1367–72. https://doi.org/10.1038/nbt.1511.
214. Orsburn BC. Proteome discoverer—A community enhanced data processing suite for protein informatics. *Proteomes* 2021;**9**:15. https://doi.org/10.3390/proteomes9010015.
215. Muth T, Behne A, Heyer R. *et al.* The MetaProteomeAnalyzer: A powerful open-source software suite for Metaproteomics data analysis and interpretation. *J Proteome Res* 2015;**14**:1557–65. https://doi.org/10.1021/pr501246w.
216. Kong AT, Leprevost FV, Avtonomov DM. *et al.* MSFragger: Ultrafast and comprehensive peptide identification in mass spectrometry–based proteomics. *Nat Methods* 2017;**14**:513–20. https://doi.org/10.1038/nmeth.4256.
217. Tyanova S, Temu T, Sinitcyn P. *et al.* The Perseus computational platform for comprehensive analysis of (prote)omics data. *Nat Methods* 2016;**13**:731–40. https://doi.org/10.1038/nmeth.3901.
218. Choi M, Chang C-Y, Clough T. *et al.* MSstats: An R package for statistical analysis of quantitative mass spectrometry-based proteomic experiments. *Bioinformatics* 2014;**30**:2524–6. https://doi.org/10.1093/bioinformatics/btu305.
219. Easterly CW, Sajulga R, Mehta S. *et al.* metaQuantome: An integrated, quantitative Metaproteomics approach reveals connections between taxonomy and protein function in complex microbiomes ∗. *Mol Cell Proteomics* 2019;**18**:S82–91. https://doi.org/10.1074/mcp.RA118.001240.
220. Sinitcyn P, Hamzeiy H, Salinas Soto F. *et al.* MaxDIA enables library-based and library-free data-independent acquisition proteomics. *Nat Biotechnol* 2021;**39**:1563–73. https://doi.org/10.1038/s41587-021-00968-7.
221. Demichev V, Messner CB, Vernardis SI. *et al.* DIA-NN: Neural networks and interference correction enable deep proteome coverage in high throughput. *Nat Methods* 2020;**17**:41–4. https://doi.org/10.1038/s41592-019-0638-x.
222. Schmid R, Heuckeroth S, Korf A. *et al.* Integrative analysis of multimodal mass spectrometry data in MZmine 3. *Nat Biotechnol* 2023;**41**:447–9. https://doi.org/10.1038/s41587-023-01690-2.
223. Smith CA, Want EJ, O'Maille G. *et al.* XCMS: Processing mass spectrometry data for metabolite profiling using nonlinear peak alignment, matching, and identification. *Anal Chem* 2006;**78**:779–87. https://doi.org/10.1021/ac051437y.
224. Wang M, Carver JJ, Phelan VV. *et al.* Sharing and community curation of mass spectrometry data with global natural products social molecular networking. *Nat Biotechnol* 2016;**34**:828–37.
225. Ruttkies C, Schymanski EL, Wolf S. *et al.* MetFrag relaunched: Incorporating strategies beyond in silico fragmentation. *J Chem* 2016;**8**:3. https://doi.org/10.1186/s13321-016-0115-9.
226. Tsugawa H, Kind T, Nakabayashi R. *et al.* Hydrogen rearrangement rules: Computational MS/MS fragmentation and structure elucidation using MS-FINDER software. *Anal Chem* 2016;**88**:7946–58. https://doi.org/10.1021/acs.analchem.6b00770.
227. Li S, Park Y, Duraisingham S. *et al.* Predicting network activity from high throughput metabolomics. *PLoS Comput Biol* 2013;**9**:e1003123. https://doi.org/10.1371/journal.pcbi.1003123.
228. Rohart F, Gautier B, Singh A. *et al.* mixOmics: An R package for 'omics feature selection and multiple data integration. *PLoS Comput Biol* 2017;**13**:e1005752. https://doi.org/10.1371/journal.pcbi.1005752.
229. Argelaguet R, Arnol D, Bredikhin D. *et al.* MOFA+: A statistical framework for comprehensive integration of multi-modal single-cell data. *Genome Biol* 2020;**21**:111.
230. Xia J, Psychogios N, Young N. *et al.* MetaboAnalyst: A web server for metabolomic data analysis and interpretation. *Nucleic Acids Res* 2009;**37**:W652–60. https://doi.org/10.1093/nar/gkp356.
231. Zhou G, Xia J. OmicsNet: A web-based tool for creation and visual analysis of biological networks in 3D space. *Nucleic Acids Res* 2018;**46**:W514–22. https://doi.org/10.1093/nar/gky510.
232. Pfeuffer J, Bielow C, Wein S. *et al.* OpenMS 3 enables reproducible analysis of large-scale mass spectrometry data. *Nat Methods* 2024;**21**:365–7. https://doi.org/10.1038/s41592-024-02197-7.
233. Borenstein E. Computational systems biology and in silico modeling of the human microbiome. *Brief Bioinform* 2012;**13**:769–80. https://doi.org/10.1093/bib/bbs022.
234. Costello EK, Stagaman K, Dethlefsen L. *et al.* The application of ecological theory toward an understanding of the human microbiome. *Science* 2012;**336**:1255–62. https://doi.org/10.1126/science.1224203.
235. Li S, Lam J, Souliotis L. *et al.* Posttranscriptional regulation in response to different environmental stresses in campylobacter jejuni. *Microbiol Spectr* 2022;**10**:e00203–22.
236. Daybog I, Kolodny O. A computational framework for resolving the microbiome diversity conundrum. *Nat Commun* 2023;**14**:7977. https://doi.org/10.1038/s41467-023-42768-4.
237. Coyte KZ, Schluter J, Foster KR. The ecology of the microbiome: Networks, competition, and stability. *Science* 2015;**350**:663–6. https://doi.org/10.1126/science.aad2602.
238. Ponomarova O, Gabrielli N, Sévin DC. *et al.* Yeast creates a niche for symbiotic lactic acid bacteria through nitrogen overflow. *Cell Syst* 2017;**5**:345–357.e6. https://doi.org/10.1016/j.cels.2017.09.002.

239. Hoegenauer C, Hammer HF, Mahnert A. *et al.* Methanogenic archaea in the human gastrointestinal tract. *Nat Rev Gastroenterol Hepatol* 2022;**19**:805–13. https://doi.org/10.1038/s41575-022-00673-z.
240. Correia-Melo C, Kamrad S, Tengölics R. *et al.* Cell-cell metabolite exchange creates a pro-survival metabolic environment that extends lifespan. *Cell* 2023;**186**:63–79.e21. https://doi.org/10.1016/j.cell.2022.12.007.
241. Yu JSL, Correia-Melo C, Zorrilla F. *et al.* Microbial communities form rich extracellular metabolomes that foster metabolic interactions and promote drug tolerance. *Nat Microbiol* 2022;**7**:542–55. https://doi.org/10.1038/s41564-022-01072-5.
242. Machado D, Maistrenko OM, Andrejev S. *et al.* Polarization of microbial communities between competitive and cooperative metabolism. *Nat Ecol Evol* 2021;**5**:195–203. https://doi.org/10.1038/s41559-020-01353-4.
243. Wang S, Mu L, Yu C. *et al.* Microbial collaborations and conflicts: Unraveling interactions in the gut ecosystem. *Gut Microbes* 2024;**16**:2296603. https://doi.org/10.1080/19490976.2023.2296603.
244. Nedialkova LP, Denzler R, Koeppel MB. *et al.* Inflammation fuels colicin Ib-dependent competition of salmonella serovar typhimurium and E. Coli in enterobacterial blooms. *PLoS Pathog* 2014;**10**:e1003844. https://doi.org/10.1371/journal.ppat.1003844.
245. Yung DBY, Sircombe KJ, Pletzer D. Friends or enemies? The complicated relationship between Pseudomonas aeruginosa and Staphylococcus aureus. *Mol Microbiol* 2021;**116**:1–15. https://doi.org/10.1111/mmi.14699.
246. García-Bayona L, Comstock LE. Bacterial antagonism in host-associated microbial communities. *Science* 2018;**361**:eaat2456. https://doi.org/10.1126/science.aat2456.
247. Good BH, Rosenfeld LB. Eco-evolutionary feedbacks in the human gut microbiome. *Nat Commun* 2023;**14**:7146.
248. Faust K, Sathirapongsasuti JJ, Izard J. *et al.* Microbial co-occurrence relationships in the human microbiome. *PLoS Comput Biol* 2012;**8**:e1002606. https://doi.org/10.1371/journal.pcbi.1002606.
249. Faust K, Raes J. Microbial interactions: From networks to models. *Nat Rev Microbiol* 2012;**10**:538–50. https://doi.org/10.1038/nrmicro2832.
250. Berry D, Widder S. Deciphering microbial interactions and detecting keystone species with co-occurrence networks. *Front Microbiol* 2014;**5**:219. https://doi.org/10.3389/fmicb.2014.00219.
251. Skwara A, Gowda K, Yousef M. *et al.* Statistically learning the functional landscape of microbial communities. *Nat Ecol Evol* 2023;**7**:1823–33. https://doi.org/10.1038/s41559-023-02197-4.
252. Liu C, Li C, Jiang Y. *et al.* A guide for comparing microbial co-occurrence networks. *iMeta* 2023;**2**:e71. https://doi.org/10.1002/imt2.71.
253. Liu Z, Ma A, Mathé E. *et al.* Network analyses in microbiome based on high-throughput multi-omics data. *Brief Bioinform* 2021;**22**:1639–55. https://doi.org/10.1093/bib/bbaa005.
254. Karimian E, Motamedian E. ACBM: An integrated agent and constraint based Modeling framework for simulation of microbial communities. *Sci Rep* 2020;**10**:8695. https://doi.org/10.1038/s41598-020-65659-w.
255. Fernex D, Noack BR, Semaan R. Cluster-based network modeling—From snapshots to complex dynamical systems. *Sci Adv* 2021;**7**:eabf5006. https://doi.org/10.1126/sciadv.abf5006.
256. Xiao Y, Angulo MT, Friedman J. *et al.* Mapping the ecological networks of microbial communities. *Nat Commun* 2017;**8**:2042. https://doi.org/10.1038/s41467-017-02090-2.
257. Kodera SM, Das P, Gilbert JA. *et al.* Conceptual strategies for characterizing interactions in microbial communities. *iScience* 2022;**25**:103775.
258. Shahin M, Ji B, Dixit PD. EMBED: Essential MicroBiomE dynamics, a dimensionality reduction approach for longitudinal microbiome studies. *Npj Syst Biol Appl* 2023;**9**:1–11.
259. Kodikara S, Lê Cao K-A. Microbial network inference for longitudinal microbiome studies with LUPINE. *Microbiome* 2025;**13**:1–32.
260. Ovaskainen O, Roy DB, Fox R. *et al.* Uncovering hidden spatial structure in species communities with spatially explicit joint species distribution models. *Methods Ecol Evol* 2016;**7**:428–36. https://doi.org/10.1111/2041-210X.12502.
261. Ovaskainen O, Tikhonov G, Norberg A. *et al.* How to make more out of community data? A conceptual framework and its implementation as models and software. *Ecol Lett* 2017;**20**:561–76. https://doi.org/10.1111/ele.12757.
262. Tikhonov G, Opedal ØH, Abrego N. *et al.* Joint species distribution modelling with the r-package Hmsc. *Methods Ecol Evol* 2020;**11**:442–7. https://doi.org/10.1111/2041-210X.13345.
263. Samarasekara CL, Flint I, Wang Y. Modelling the spatial dependence of multi-species point patterns. *Ecol Evol* 2025;**15**:e71066. https://doi.org/10.1002/ece3.71066.
264. Kurtz ZD, Müller CL, Miraldi ER. *et al.* Sparse and compositionally robust inference of microbial ecological networks. *PLoS Comput Biol* 2015;**11**:e1004226. https://doi.org/10.1371/journal.pcbi.1004226.
265. Tackmann J, Matias Rodrigues JF, Von Mering C. Rapid inference of direct interactions in large-scale ecological networks from heterogeneous microbial sequencing data. *Cell Syst* 2019;**9**:286–296.e8. https://doi.org/10.1016/j.cels.2019.08.002.
266. Faust K, Raes J. *CoNet App: Inference of Biological Association Networks Using Cytoscape* 2016, F1000Res 2016;**5**:1519. https://doi.org/10.12688/f1000research.9050.2.
267. Van Den Berg NI, Machado D, Santos S. *et al.* Ecological modelling approaches for predicting emergent properties in microbial communities. *Nat Ecol Evol* 2022;**6**:855–65. https://doi.org/10.1038/s41559-022-01746-7.
268. Monaco H, Liu KS, Sereno T. *et al.* Spatial-temporal dynamics of a microbial cooperative behavior resistant to cheating. *Nat Commun* 2022;**13**:721. https://doi.org/10.1038/s41467-022-28321-9.
269. Silverstein MR, Bhatnagar JM, Segrè D. Metabolic complexity drives divergence in microbial communities. *Nat Ecol Evol* 2024;**8**:1493–504. https://doi.org/10.1038/s41559-024-02440-6.
270. Silverman JD, Washburne AD, Mukherjee S. *et al.* A phylogenetic transform enhances analysis of compositional microbiota data. *elife* 2017;**6**:e21887. https://doi.org/10.7554/eLife.21887.
271. Asnicar F, Berry SE, Valdes AM. *et al.* Microbiome connections with host metabolism and habitual diet from 1,098 deeply phenotyped individuals. *Nat Med* 2021;**27**:321–32. https://doi.org/10.1038/s41591-020-01183-8.
272. Colarusso AV, Goodchild-Michelman I, Rayle M. *et al.* Computational modeling of metabolism in microbial communities on a genome-scale. *Curr Opin Syst Biol* 2021;**26**:46–57. https://doi.org/10.1016/j.coisb.2021.04.001.
273. Mardinoglu A, Palsson BØ. Genome-scale models in human metabologenomics. *Nat Rev Genet* 2025;**26**:123–40. https://doi.org/10.1038/s41576-024-00768-0.
274. Thiele I, Palsson BØ. A protocol for generating a high-quality genome-scale metabolic reconstruction. *Nat Protoc* 2010;**5**:93–121. https://doi.org/10.1038/nprot.2009.203.

275. Magnúsdóttir S. Generation of genome-scale metabolic reconstructions for 773 members of the human gut microbiota. *Nat Biotechnol* 2017;**35**:81–9. https://doi.org/10.1038/nbt.3703.
276. Heinken A. Genome-scale metabolic reconstruction of 7,302 human microorganisms for personalized medicine. *Nat Biotechnol* 2023;**41**:1320–31. https://doi.org/10.1038/s41587-022-01628-0.
277. Greenblum S, Turnbaugh PJ, Borenstein E. Metagenomic systems biology of the human gut microbiome reveals topological shifts associated with obesity and inflammatory bowel disease. *Proc Natl Acad Sci* 2012;**109**:594–9. https://doi.org/10.1073/pnas.1116053109.
278. Heinken A, Basile A, Hertel J. *et al.* Genome-scale metabolic Modeling of the human microbiome in the era of personalized medicine. *Ann Rev Microbiol* 2021;**75**:199–222. https://doi.org/10.1146/annurev-micro-060221-012134.
279. Neal M, Thiruppathy D, Zengler K. Genome-scale metabolic modeling of the human gut bacterium Bacteroides fragilis strain 638R. *PLoS Comput Biol* 2023;**19**:e1011594. https://doi.org/10.1371/journal.pcbi.1011594.
280. Henson MA, Phalak P. Microbiota dysbiosis in inflammatory bowel diseases: in silico investigation of the oxygen hypothesis. *BMC Syst Biol* 2017;**11**:145. https://doi.org/10.1186/s12918-017-0522-1.
281. Fernandes P, Sharma Y, Zulqarnain F. *et al.* Identifying metabolic shifts in Crohn's disease using 'omics-driven contextualized computational metabolic network models. *Sci Rep* 2023;**13**:203. https://doi.org/10.1038/s41598-022-26816-5.
282. Henson MA. Interrogation of the perturbed gut microbiota in gouty arthritis patients through in silico metabolic modeling. *Eng Life Sci* 2021;**21**:489–501. https://doi.org/10.1002/elsc.202100003.
283. Shoaie S, Ghaffari P, Kovatcheva-Datchary P. *et al.* Quantifying diet-induced metabolic changes of the human gut microbiome. *Cell Metab* 2015;**22**:320–31. https://doi.org/10.1016/j.cmet.2015.07.001.
284. Zampieri G, Campanaro S, Angione C. *et al.* Metatranscriptomics-guided genome-scale metabolic modeling of microbial communities. *Cell Rep Methods* 2023;**3**:100383. https://doi.org/10.1016/j.crmeth.2022.100383.
285. Hsieh YE, Tandon K, Verbruggen H. *et al.* Comparative analysis of metabolic models of microbial communities reconstructed from automated tools and consensus approaches. *Npj Syst Biol Appl* 2024;**10**:1–10.
286. Seaver SMD. The ModelSEED biochemistry database for the integration of metabolic annotations and the reconstruction, comparison and analysis of metabolic models for plants, fungi and microbes. *Nucleic Acids Res* 2021;**49**:D575–88. https://doi.org/10.1093/nar/gkaa746.
287. Capela J, Lagoa D, Rodrigues R. *et al.* Merlin, an improved framework for the reconstruction of high-quality genome-scale metabolic models. *Nucleic Acids Res* 2022;**50**:6052–66. https://doi.org/10.1093/nar/gkac459.
288. Wang H, Marcišauskas S, Sánchez BJ. *et al.* RAVEN 2.0: A versatile toolbox for metabolic network reconstruction and a case study on Streptomyces coelicolor. *PLoS Comput Biol* 2018;**14**:e1006541. https://doi.org/10.1371/journal.pcbi.&break;1006541.
289. Machado D, Andrejev S, Tramontano M. *et al.* Fast automated reconstruction of genome-scale metabolic models for microbial species and communities. *Nucleic Acids Res* 2018;**46**:7542–53. https://doi.org/10.1093/nar/gky537.
290. Zimmermann J, Kaleta C, Waschina S. Gapseq: Informed prediction of bacterial metabolic pathways and reconstruction of accurate metabolic models. *Genome Biol* 2021;**22**:81. https://doi.org/10.1186/s13059-021-02295-1.
291. Lieven C. MEMOTE for standardized genome-scale metabolic model testing. *Nat Biotechnol* 2020;**38**:272–6. https://doi.org/10.1038/s41587-020-0477-4.
292. Heirendt L, Arreckx S, Pfau T. *et al.* Creation and analysis of biochemical constraint-based models using the COBRA toolbox v.3.0. *Nat Protoc* 2019;**14**:639–702. https://doi.org/10.1038/s41596-018-0098-2.
293. Arkin AP, Cottingham RW, Henry CS. *et al.* KBase: The United States Department of Energy Systems Biology Knowledgebase. *Nat Biotechnol* 2018;**36**:566–9. https://doi.org/10.1038/nbt.4163.
294. Lewis NE, Nagarajan H, Palsson BO. Constraining the metabolic genotype–phenotype relationship using a phylogeny of in silico methods. *Nat Rev Microbiol* 2012;**10**:291–305. https://doi.org/10.1038/nrmicro2737.
295. Royer C, Patin NV, Jesser KJ. *et al.* Comparison of metagenomic and traditional methods for diagnosis of E. Coli enteric infections. *MBio* 2024;**15**:e03422–3. https://doi.org/10.1128/mbio.03422-23.
296. Simner PJ, Miller S, Carroll KC. Understanding the promises and hurdles of metagenomic next-generation sequencing as a diagnostic tool for infectious diseases. *Clin Infect Dis* 2018;**66**:778–88. https://doi.org/10.1093/cid/cix881.
297. Jousset A, Bienhold C, Chatzinotas A. *et al.* Where less may be more: How the rare biosphere pulls ecosystems strings. *ISME J* 2017;**11**:853–62. https://doi.org/10.1038/ismej.2016.174.
298. Succurro A, Ebenhöh O. Review and perspective on mathematical modeling of microbial ecosystems. *Biochem Soc Trans* 2018;**46**:403–12. https://doi.org/10.1042/BST20170265.
299. Chakraborty D, Jousset A, Wei Z. *et al.* Rare taxa in the core microbiome. *Trends Microbiol* 2025;S0966842X25000782. https://doi.org/10.1016/j.tim.2025.03.002.
300. Tiwari K, Kananathan S, Roberts MG. *et al.* Reproducibility in systems biology modelling. *Mol Syst Biol* 2021;**17**:e9982. https://doi.org/10.15252/msb.20209982.
301. Berlo RJ. Predicting metabolic fluxes using gene expression differences as constraints. *IEEEACM Trans Comput Biol Bioinform* 2011;**8**:206–16.
302. Gatto F, Miess H, Schulze A. *et al.* Flux balance analysis predicts essential genes in clear cell renal cell carcinoma metabolism. *Sci Rep* 2015;**5**:5. https://doi.org/10.1038/srep10738.
303. Hackett SR, Zanotelli VRT, Xu W. *et al.* Systems-level analysis of mechanisms regulating yeast metabolic flux. *Science* 2016;**354**:aaf2786. https://doi.org/10.1126/science.aaf2786.
304. Herrmann HA, Dyson BC, Vass L. *et al.* Flux sampling is a powerful tool to study metabolism under changing environmental conditions. *Npj Syst Biol Appl* 2019;**5**:1–8.
305. Özcan E, Çakır T. Reconstructed metabolic network models predict flux-level metabolic reprogramming in glioblastoma. *Front Neurosci* 2016;**10**:10. https://doi.org/10.3389/fnins.2016.00156.
306. Nanda P, Ghosh A. Genome scale-differential flux analysis reveals deregulation of lung cell metabolism on SARS-CoV-2 infection. *PLoS Comput Biol* 2021;**17**:e1008860. https://doi.org/10.1371/journal.pcbi.1008860.
307. Hashemi S, Razaghi-Moghadam Z, Nikoloski Z. Identification of flux trade-offs in metabolic networks. *Sci Rep* 2021;**11**:23776. https://doi.org/10.1038/s41598-021-03224-9.

308. Moreno-Villena JJ, Zhou H, Gilman IS. *et al.* Spatial resolution of an integrated C4+CAM photosynthetic metabolism. *Sci Adv* 2022;**8**:eabn2349. https://doi.org/10.1126/sciadv.abn2349.
309. Schäfer M, Pacheco AR, Künzler R. *et al.* Metabolic interaction models recapitulate leaf microbiota ecology. *Science* 2023;**381**:eadf5121. https://doi.org/10.1126/science.adf5121.
310. Li G, Liu L, Du W. *et al.* Local flux coordination and global gene expression regulation in metabolic modeling. *Nat Commun* 2023;**14**:5700. https://doi.org/10.1038/s41467-023-41392-6.
311. Harcombe WR, Riehl WJ, Dukovski I. *et al.* Metabolic resource allocation in individual microbes determines ecosystem interactions and spatial dynamics. *Cell Rep* 2014;**7**:1104–15. https://doi.org/10.1016/j.celrep.2014.03.070.
312. Eduardo MB, Cottone G, McCloskey CW. *et al.* A metabolic shift to the serine pathway induced by lipids fosters epigenetic reprogramming in nontransformed breast cells. *Sci Adv* 2025;**11**:eads9182. https://doi.org/10.1126/sciadv.ads9182.
313. Smith EN, van Aalst M, Weber APM. *et al.* Alternatives to photorespiration: A system-level analysis reveals mechanisms of enhanced plant productivity. *Sci Adv* 2025;**11**:eadt9287. https://doi.org/10.1126/sciadv.adt9287.
314. Norsigian CJ, Fang X, Seif Y. *et al.* A workflow for generating multi-strain genome-scale metabolic models of prokaryotes. *Nat Protoc* 2020;**15**:1–14. https://doi.org/10.1038/s41596-019-0254-3.
315. Chetty A, Blekhman R. Multi-omic approaches for host-microbiome data integration. *Gut Microbes* 2024;**16**:2297860. https://doi.org/10.1080/19490976.2023.2297860.
316. Venturelli OS, Carr AV, Fisher G. *et al.* Deciphering microbial interactions in synthetic human gut microbiome communities. *Mol Syst Biol* 2018;**14**:e8157. https://doi.org/10.15252/msb.20178157.
317. Clark RL, Connors BM, Stevenson DM. *et al.* Design of synthetic human gut microbiome assembly and butyrate production. *Nat Commun* 2021;**12**:3254.
318. Ezzamouri B, Shoaie S, Ledesma-Amaro R. Synergies of systems biology and synthetic biology in human microbiome studies. *Front Microbiol* 2021;**12**:681982. https://doi.org/10.3389/fmicb.2021.681982.
319. Mousavinasab F, Karimi R, Taheri S. *et al.* Microbiome modulation in inflammatory diseases: Progress to microbiome genetic engineering. *Cancer Cell Int* 2023;**23**:271. https://doi.org/10.1186/s12935-023-03095-2.
320. Bai X, Huang Z, Duraj-Thatte AM. *et al.* Engineering the gut microbiome. *Nat Rev Bioeng* 2023;**1**:665–79. https://doi.org/10.1038/s44222-023-00072-2.
321. Arnold JW, Roach J, Azcarate-Peril MA. Emerging technologies for gut microbiome research. *Trends Microbiol* 2016;**24**:887–901. https://doi.org/10.1016/j.tim.2016.06.008.
322. Van Neerven SM, Ramadan R, Van Driel MS. *et al.* Intestinal organoid co-culture protocol to study cell competition in vitro. *STAR Protoc* 2022;**3**:101050. https://doi.org/10.1016/j.xpro.2021.101050.
323. Fofanova TY, Karandikar UC, Auchtung JM. *et al.* A novel system to culture human intestinal organoids under physiological oxygen content to study microbial-host interaction. *PLoS One* 2024;**19**:e0300666. https://doi.org/10.1371/journal.pone.0300666.
324. Pal S, Bhattacharya M, Lee S-S. *et al.* Quantum computing in the next-generation computational biology landscape: From protein folding to molecular dynamics. *Mol Biotechnol* 2024;**66**:163–78. https://doi.org/10.1007/s12033-023-00765-4.
325. Saggi MK, Bhatia AS, Isaiah M. *et al.* Multi-Omic and quantum machine learning integration for lung subtypes classification. *Future Generation Computer Systems* 2025:107905.
326. Gupta RS, Wood CE, Engstrom T. *et al.* A systematic review of quantum machine learning for digital health. *Npj Digit Med* 2025;**8**:1–15. https://doi.org/10.1038/s41746-025-01597-z.
327. Harrow AW, Montanaro A. Quantum computational supremacy. *Nature* 2017;**549**:203–9. https://doi.org/10.1038/nature23458.
328. Higgins JS, Lloyd LT, Sohail SH. *et al.* Photosynthesis tunes quantum-mechanical mixing of electronic and vibrational states to steer exciton energy transfer. *Proc Natl Acad Sci* 2021;**118**:e2018240118. https://doi.org/10.1073/pnas.2018240118.
329. Riera Aroche R, Ortiz García YM, Martínez Arellano MA. *et al.* DNA as a perfect quantum computer based on the quantum physics principles. *Sci Rep* 2024;**14**:11636. https://doi.org/10.1038/s41598-024-62539-5.
330. Biamonte J, Wittek P, Pancotti N. *et al.* Quantum machine learning. *Nature* 2017;**549**:195–202. https://doi.org/10.1038/nature23474.
331. Marais A, Adams B, Ringsmuth AK. *et al.* The future of quantum biology. *J R Soc Interface* 2018;**15**:20180640. https://doi.org/10.1098/rsif.2018.0640.
332. Islam N, Pal S, Misra S. QBaN: Quantum bacterial nanonetworks for secure molecular communication. *IEEE Trans Mol Biol Multi-Scale Commun* 2024;**10**:633–41. https://doi.org/10.1109/TMBMC.2024.3476192.

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