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REVIEW

Diet and the Human Gut Microbiome: An International Review

Annette S. Wilson[1] · Kathryn R. Koller[2] · Matsepo C. Ramaboli[3] · Lucky T. Nesengani[3] · Soeren Ocvirk[1,4] · Caixia Chen[1] · Christie A. Flanagan[2] · Flora R. Sapp[2] · Zoe T. Merritt[2] · Faheem Bhatti[1] · Timothy K. Thomas[2] · Stephen J. D. O'Keefe[1,3]

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Abstract

This review summarizes the key results of recently published studies on the effects of dietary change and nutritional intervention on the human microbiome from around the world, focusing on the USA, Canada, Europe, Asia, and Africa. It first explores mechanisms that might explain the ability of fiber-rich foods to suppress the incidence and mortality from westernized diseases, notably cancers of the colon, breast, liver, cardiovascular, infectious, and respiratory diseases, diabetes, and obesity (O'Keefe in Lancet Gastroenterol Hepatol 4(12):984–996, 2019; Am J Clin Nutr 110:265–266, 2019). It summarizes studies from Africa which suggest that disturbance of the colonic microbiome may exacerbate chronic malnutrition and growth failure in impoverished communities and highlights the importance of breast feeding. The American section discusses the role of the microbiome in the swelling population of patients with obesity and type 2 diabetes and examines the effects of race, ethnicity, geography, and climate on microbial diversity and metabolism. The studies from Europe and Asia extoll the benefits of whole foods and plant-based diets. The Asian studies examine the worrying changes from low-fat, high-carbohydrate diets to high-fat, low-carbohydrate ones and the increasing appearance of westernized diseases as in Africa and documents the ability of high-fiber traditional Chinese diets to reverse type 2 diabetes and control weight loss. In conclusion, most of the studies reviewed demonstrate clear changes in microbe abundances and in the production of fermentation products, such as short-chain fatty acids and phytochemicals following dietary change, but the significance of the microbiota changes to human health, with the possible exception of the stimulation of butyrogenic taxa by fiber-rich foods, is generally implied and not measured. Further studies are needed to determine how these changes in microbiota composition and metabolism can improve our health and be used to prevent and treat disease.

Keywords Diet · Microbiome · Metabolome · Fiber · Fat · Colon cancer · Obesity · Type 2 diabetes · Westernized diseases · Prebiotics · Probiotics · Short-chain fatty acids · Bile acids

✉ Stephen J. D. O'Keefe
sjokeefe@pitt.edu

[1] Division of Gastroenterology, Hepatology and Nutrition, Department of Medicine, University of Pittsburgh, W1145 Biomedical Science Tower, 200 Lothrop Street, Pittsburgh, PA 15213, USA

[2] Clinical and Research Services, Community Health Services, Alaska Native Tribal Health Consortium, Anchorage, AK, USA

[3] African Microbiome Institute, Stellenbosch University, Cape Town, Western Cape, South Africa

[4] Department of Gastrointestinal Microbiology, German Institute of Human Nutrition Potsdam-Rehbruecke, Nuthetal, Germany

Annette S. Wilson

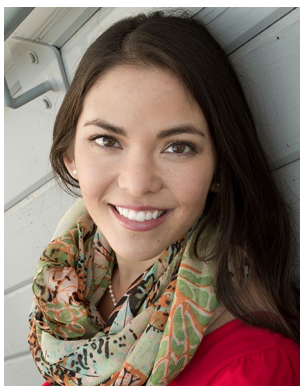
Kathryn R. Koller

Soeren Ocvirk

Matsepo C. Ramaboli

Caixia Chen

Lucky T. Nesengani



Christie A. Flanagan

Flora R. Sapp

Timothy K. Thomas

Zoe T. Merritt

Stephen J. D. O'Keefe

Faheem Bhatti

Source and Selection of Articles Reviewed

The aim of this review is to summarize recently published research from human studies on the gut microbiota-modulating effects of diet. It includes sections on microbiome research of populations from around the globe in the USA, Canada, Europe, Asia, and Africa that address specific public health challenges. References for this review were identified through searches of PubMed for peer-reviewed articles published in English from 2014 to August 31, 2019 by use of the terms "Human Nutrition (or Nutrition) and the (Human) Microbiome in USA, Canada, Europe, Asia, and Africa," appearing in the title or abstract. Articles resulting from these searches, as well as relevant references cited in those articles, were reviewed. The conclusions highlight the future needs and implications for scientists and clinicians in this fast-developing field of research, and the need for high-quality, large-scale controlled human dietary intervention studies.

Human Gut Microbiome and Its Metabolites in Health and Disease

The human gut contains trillions of bacterial cells that are reported to be at a ratio of approximately 1:1 with our own cells [1]. Thus, they form a vibrant living population that has a metabolic activity similar to the liver. There is consequently a move to describe the microbiota as a whole a new human organ. Their intimate association with the intestinal mucosa, which is also highly biologically active with a high cellular turnover rate, can be expected to have a major impact on human health and the prevention or precipitation of disease. The gut microbiota has a symbiotic relationship with the host, playing a role in maintaining health and metabolic homeostasis through the production of many metabolites. The collective genomes of these microorganisms that inhabit the gut refer to the metagenome. The development of the gut microbiome begins at birth and in healthy individuals is largely completed within 3 years, but can be modified by environmental factors, particularly diet composition and volume, and antibiotic therapy [2]. This review will focus on the modulatory effects of diet.

Research on the microbiome has expanded rapidly worldwide providing novel insights into both human and animal diseases and interventions designed to alleviate them [3, 4].

Bacterial populations inhabit the entire length of the human gut from the oral cavity to the rectum. The density and composition of the resident microbial communities, however, differ markedly between anatomical sites and depend on transit rates, host secretions, environmental conditions, substrate availability, and the integrity of the gut wall [5]. Overall the predominant bacterial groups in the microbiome are gram-positive Firmicutes and gram-negative Bacteroidetes [6]. The bacteria that reside in the distal parts of the gut contribute to host health through biosynthesis of vitamins and essential amino acids and by production of important metabolites from nondigestible food components [7]. Short-chain fatty acid (SCFA) metabolites, notably butyrate, propionate and acetate are the by-products of bacterial fermentation of indigestible carbohydrates, mostly fiber, and act as the major energy source for the colonic epithelial cells and therefore strengthen the mucosal barrier and defense [8].

Dietary Fiber and the Microbiome

The microbiota is dependent on food residues for survival and metabolism. Recent reviews by ourselves [9] have shown that the fermentation of carbohydrate residues, namely fiber, releases metabolites that not only provide food for the colonic epithelium, but also exert a remarkable variety of regulatory effects on colonic mucosal inflammation and proliferation. This helps explain the epidemiological studies from throughout the world that have shown that high fiber consumption maintains colonic health and prevents colon cancer [9]. Furthermore, high fiber intakes provide high rates of butyrogenesis, which exceed the metabolic requirements of the colonic mucosa and enter the blood stream to exert epigenetic and immunomodulatory effects on other organs in the body (Fig. 1). This, in turn, explains the association between high fiber intakes and a reduction in incidence and mortality from all westernized diseases, including cancers of the colon, breast, liver [10, 11], cardiovascular, infectious, and respiratory diseases, diabetes, and obesity [12–14]. Similar conclusions were drawn by Reynolds based on nearly 135 million person-years of data from 185 prospective studies and 58 clinical trials with 4635 adult participants comparing the highest dietary fiber consumers with the lowest. Here, they found a 15–30% lower rate in all-cause and cardiovascular-related mortality, in addition to a lower incidence and mortality in colorectal and breast cancer, coronary heart disease, stroke, and type 2 diabetes. These studies were recently quoted by O'Keefe in his viewpoint for Lancet GI [10] where he updated the evidence supporting Burkitts fiber hypothesis that the deficiency of fiber in the westernized diet can explain much of the morbidity and mortality associated with westernized diseases.

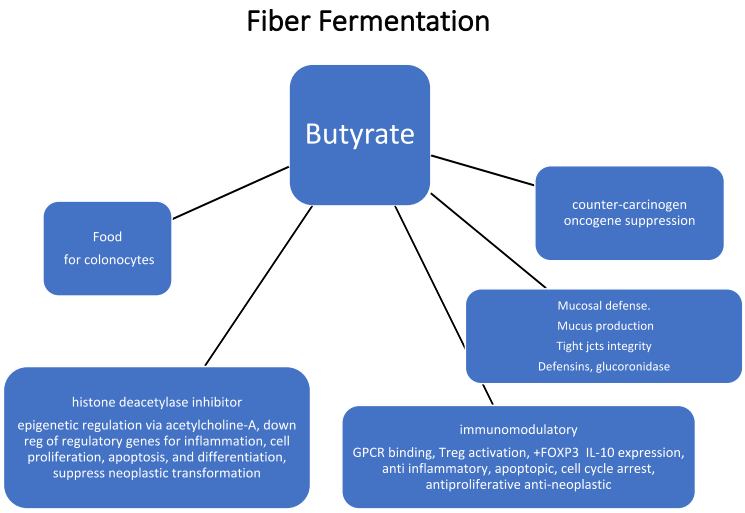


Fig. 1 Illustration of some of the actions of butyrate released by colonic microbial fermentation of fiber on host metabolism, inflammation, and carcinogenesis

Nutrition and the Gut Microbiome in Africa

Diseases of undernutrition and overnutrition that pose widespread health challenges in Africa are viewed as being "microbiota-dependent," and hence, their mitigation would benefit from "microbiota-directed" interventions [15]. Understanding the role of the microbiota in their pathogenesis is indispensable in informing intervention strategies. Research of the past 5 years on human microbiome and nutrition in Africa, as reviewed in this section, is dominated by intervention trials of childhood undernutrition and exploration of the effect of urbanization on the rising incidences of westernized diseases. In general, studies on undernutrition (summarized in Table 1) investigate impairments of the developing gut microbiome associated with malnutrition and its treatment sequelae while research on westernized diseases aims at finding how the changes in diet and lifestyle affect the gut microbiome and contribute to the increasing risk of these diseases in communities where the incidences were historically low.

Undernutrition and the Gut Microbiome

Undernutrition is a global health challenge accounting for 45% of childhood mortality and higher morbidities in sub-Saharan Africa and South Asia [16]. Malnutrition is a complex of disorders arising from inadequate nutrition that include gut microbiota dysbiosis, metabolic, immune and intestinal barrier dysfunction, as well as micronutrient deficiencies. It is often exacerbated by enteropathogenic and HIV infections, enteropathy, diarrhea, and chronic inflammation. It manifests as severe or moderate, acute or chronic, edematous or nonedematous forms, all of which are managed through hospitalization and subsequent nutritional rehabilitation coupled with antibiotic therapy [17]. The challenge in Africa is that, despite the implementation of the World Health Organization (WHO)-recommended treatment protocol, there is still high mortality among children undergoing treatment. It is thought that severe acute malnutrition (SAM) is complicated by co-occurrence in a vicious cycle with infections culminating in death [18]. Malnutrition results in adverse health outcomes such as stunting, immune dysfunction, and cognitive impairments [15]. In 2011, Gordon and colleagues proposed a hypothetical model of relationships between the gut microbiota, the immune system, and diet, which underlie the development of malnutrition [19]. Years later, they provided evidence that gut microbiota dysbiosis is causally linked to childhood undernutrition, based on longitudinal studies involving Malawian infants and children with varying degrees of undernutrition and corresponding growth rates of gnotobiotic mice to which the kids' fecal samples had been transplanted [15, 20]. Since then, several intervention and observational studies [21], completed (Table 1) [22–32] and ongoing [33], have sought to understand the role of gut microbiota in nutritional outcomes, childhood morbidities, and enteropathy associated with malnutrition in limited resources settings to improve outcomes. Vray and associates embarked on alternative "renutrition" strategies for the management of moderate acute malnutrition among children across Madagascar, Niger, Central African Republic, and Senegal [34].

Iron-containing micronutrients powders (MNP) are widely used in sub-Saharan Africa in the prevention of iron deficiency anemia and micronutrient deficiencies in infants [23]. However, there is concern that their iron content may have adverse effects in children simultaneously treated with antibiotics. Antibiotic use is remarkably

Table 1 Studies of the gut microbiome in undernourished children in Africa from 2015 to 2019

Location of study and reference	Disease or condition	Age of participants	Number and/or nutritional status of participants	Study design	Study outcomes
Uganda Atukunda et al. 2019	Undernutrition	20–24 months	$n=77$ intervention; $n=78$ control	Follow-up of open cluster-randomized intervention trial	Cognitive development, growth, gut microbiota
Kenya Paganini et al. 2019	Diarrhea and iron deficiency anemia	8–10 months	$n=28$: 4 groups (with and without iron and antibiotics)	Sub-study of a double-blind randomized controlled intervention trial	Gut microbiome, enteropathogens, gut inflammation, fecal pH, morbidity
Kenya Tang et al. 2017	Iron deficiency and iron deficiency anemia	6 months	$n=45$: 3 groups (with and without iron and placebo)	Double-blind individually randomized controlled trial	Gut microbiome, gut inflammation, zinc absorption
Gambia Davis et al. 2017	Undernutrition	1-5 months	33 mother/infant pairs at 4, 16, and 20 weeks postpartum	Sub-study of a randomized trial	Human milk oligosaccharides (HMO), infant gut microbiota, gut inflammation, growth, morbidity
Malawi Ordiz et al. 2017	Environmental enteric dysfunction (EED)	12–23 months	$n=81$; 47 boys and 34 girls	Legume intervention trial	Gut microbiota, gut absorption, morbidity
Niger and *Senegal* Million et al. 2016	SAM	< 60 months	$n=86$; not in feeding programs; prior to supplements	Two prospective case–control studies	Gut microbiota, aerotolerant odds ratio, gut redox potential, *Methanobrevibacter smithii*
Malawi Charbonneau et al. 2016	Undernutrition	6 months	$n=29$ healthy; $n=59$ severe stunting	Sub-study of two randomized controlled single-blind parallel group trials, germ-free mouse and piglet studies	HMO composition, infant gut microbiota, growth
Uganda Kristensen et al. 2016	SAM	6–24 months	$n=87$: all hospitalized with SAM	Sub-study of an observational study	Gut microbiota, nutritional outcomes
Malawi Reyes et al. 2015	SAM	0–30 months	Eight monozygotic healthy twin pairs; 12 dizygotic twin pairs (six twin pairs discordant for kwashiorkor; and the other 6 discordant for marasmus)	Longitudinal studies, follow-up of the Malawian discordant twin trial Germ-free mouse study	Gut microbiota DNA viromes, nutritional outcomes
Malawi Dewey et al. 2015	Diet-dependent enteropathy in SAM	21 months	One monozygotic twin pair discordant for kwashiorkor	Longitudinal studies, follow-up of the Malawian discordant twin trial Germ-free mouse study	IgA^+ bacterial taxa of the fecal microbiota, weight loss, sepsis
Kenya Jaeggi et al. 2015	Iron deficiency and iron deficiency anemia	6 months	$n=115$, consuming home-fortified maize porridge with or without iron	Two double-blind randomized controlled trials	Gut microbiome, gut inflammation, morbidity

common in impoverished communities in Kenya, with an average of 4.9 antibiotic courses per child year. In rural Tanzania, more than 50% of infants receive antibiotics by the age of 6 months, chiefly for diarrheal and respiratory diseases. In a controlled intervention trial, Paganini et al. gave small groups (*n* 6–8) healthy infants who were still being breast-fed various combinations of MNP with and without iron (5 mg), and with and without antibiotics (amoxicillin 125 mg/day; ampicillin 125 mg/day + cloxacillin 125 mg/day; or trimethoprim 40 mg/day + sulfamethoxazole 200 mg/day). They were concerned to find that iron supplementation increased the appearance of pathogens such as pathogenic *E. coli* and decreased *Bifidobacterium* abundance. In other separate studies Jaeggi et al. and Tang et al. made similar observations that iron supplementation increased enteropathogen levels and intestinal inflammation in weaning infants [24, 25]. Notably, a randomized clinical trial investigating the effect of antibiotics on the gut microbiome diversity in Burkina Faso preschool children also reported that the use of Azithromycin affected the composition of the intestinal microbiome [22]. Although these findings discourage use of iron-containing micronutrient powders during antibiotic therapy, they raise concern over the empiric use of antibiotics and nutritional supplements in impoverished communities, where more attention should arguably be paid to improving food, sanitation, and housing.

Malawi has exceedingly high rates of infant mortality from malnutrition [35]. Consequently, there is a desperate need for developing efficacious interventions to ameliorate SAM. To investigate this, Malawian infant twin pairs discordant and concordant for marasmus and kwashiorkor have been involved as microbiota donors in a series of longitudinal studies designed to understand underlying mechanisms of SAM [20, 31, 32]. Remarkably, undernutrition-associated phenotypes such as stunting [29] and diet-dependent enteropathy [32] could be transferred to gnotobiotic mouse models through transplantation of fecal samples or culture-purified bacteria obtained from sick infants. Bearing in mind the previously described low efficacy of ready-to-use therapeutic foods (RUTFs) [20], there is hope that identification of distinct microbial biomarkers that characterize different states of malnutrition, such as those indicative of stunting and enteropathy, as well as those that can ameliorate these pathologies, is indispensable in evaluating interventions, in different socioeconomic settings and geographic locations [2, 32]. Indeed, the gut microbiota maturity, defined using bacterial taxonomic biomarkers, discriminatory for age and predictive for growth, has been described [2]. Its potential as a lead in studying the effects of host and environmental factors on the gut microbiota development and associated diseases has been demonstrated [15, 32]. Such studies portray the coupling of infant twin microbiota donors with gnotobiotic mouse models as powerful tools in investigating the role of gut microbiome in childhood undernutrition and its interventions.

Breast milk is a source of nutrients and bioactive substances required for normal growth and development of the infant [36]. It contains pro-, and prebiotics such as milk oligosaccharides [37] that are viewed as potential microbiota-directed therapeutics for childhood undernutrition. These oligosaccharides are beneficial substrates to the gut microbiota contributing to enhanced gut bacteria function, protection from pathogen infection, and improved immune response. Gordon and co-workers analyzed human milk oligosaccharides (HMOs) from breast milk of 6-month postpartum mothers in two independent randomized controlled clinical trials of undernourishment in Malawi [29]. The first birth cohort study tested the efficacy of lipid-based nutrient supplements (LNS) to prevent severe stunting among infants (LCNI-5 study), while the second investigated supplementing maternal and infant diet with micronutrient-fortified LNS (iLiNS-DYAD-M study).

Initially, breast milk samples were selected from mothers in the LCNI-5 study whose children exhibited healthy growth (height-for-age Z score [HAZ] > 0; $n = 29$) or severe stunting (HAZ < −3; $n = 59$). The HMO abundance and composition were found to be associated with infant growth since the breast milk from mothers whose children were severely stunted had significantly lower levels of total, sialylated, and fucosylated HMOs compared to those from mothers whose infants had healthy growth. Among the mothers lacking the *FUT2* gene, i.e., unable to produce alpha-1,2-linked fucosylated HMOs ("nonsecretor" phenotype), those whose infants were severely stunted produced milk HMOs deficient in fucosylated and sialylated components compared to milk HMOs from mothers of healthy infants. Similar observations were made in mothers enrolled in the iLiNS-DYAD-M study. However, in "secretor" mothers in the latter study, only the sialylated milk HMOs, neither total nor fucosylated, were elevated significantly in those with healthy infants ($n = 20$) compared to the ones with stunted infants ($n = 40$).

To investigate the role of sialylated milk HMOs in infant growth, the researchers used bacteria from fecal microbiota of a stunted infant donor to colonize germ-free mice and piglets feeding on prototypic Malawian diet (control) selectively supplemented with sialylated bovine milk oligosaccharides (S-BMOs) or inulin. Their objective was to determine preclinically whether the sialylated HMOs are causally related to healthy growth. After colonizing 5-week-old male germ-free mice with a defined community of 25 bacterial strains, and monitoring body weight and composition for 5 weeks, weight and lean body mass gain were significantly increased by S-BMO supplementation and not inulin. Of the 25 bacterial strains, only *Escherichia coli* and *Bacteroides fragilis* responded transcriptionally to S-BMO supplementation

in vivo; however, interaction with all the community members was necessary to mediate growth promotion. Further, observations of decreased levels of serum and liver acylcarnitines and increased concentrations of serum insulin, leptin, and triglycerides in S-BMO-treated mice compared to controls, over the initial 5-week period, suggested that S-BMO supplementation modulated metabolism in host tissues. Overall, this study demonstrated the causal relationship between S-BMOs and growth as well as the potential of S-BMOs as supplements that have to be tested for improving growth outcomes. In a randomized trial, Davis et al. [26] investigated the relationships between HMOs, microbiota, infant morbidity, and growth in Gambian mother–infant dyads. Without providing causation mechanisms, the fucosylated HMOs were found to be associated with morbidity while sialylated HMOs were predictive of growth. *Prevotella* was associated with decreased morbidity, as had been observed in a previous Malawian children cohort study cited [38].

Westernized Diseases and the Gut Microbiome

Nutrition and the gut microbiome have been implicated in the etiology of many westernized diseases [9, 39]. However, the role of the gut microbiome in the rising incidences of colorectal cancer (CRC) in countries undergoing dietary and lifestyle transitions, typical of sub-Saharan Africa, is just unfolding. The age-standardized incidence of CRC per 100,000 has been traditionally < 5 [40], but, despite variations between African countries, larger incidences of > 10 have been reported in some countries [41]. In Zimbabwe, an observational study was recently undertaken to assess the link between CRC risk and dietary changes from traditional to Western during urbanization [41]. Owing to retention of fiber intake in the urban population, the levels of butyrate and butyrogenic bacteria were similar between rural and urban participants, yet the levels of bile acids indicative of high-fat diet were higher among urban participants. This study demonstrated the potential to prevent a CRC epidemic through retention of traditional high-fiber foods, a sustained adjustment of diet that would reverse the pattern of metabolic markers associated with CRC risk.

In a previous comparative diet switch study of rural Africans to a high-fat, low-fiber diet, and African Americans to a high-fiber, low-fat diet, reciprocal changes in microbiota, its metabolic activity, and mucosal biomarkers of CRC risk were observed within 2 weeks [42]. In yet another comparative metagenomic study, not related to a disease, but attempting to understand how the microbiome has evolved to support hosts living in different environments, the gut microbiome of the Hadza hunter-gatherers of Tanzania was compared with that of the urban Italians [43]. The Hadza gut microbiome was enriched with genes with the functional capacity to ferment carbohydrates into short-chain fatty acids (SCFAs) and synthesize SCFAs from amino acids, while the urban Italian gut microbiome showed functional potential in xenobiotics metabolism [43, 44]. It has been assessed that the current diet of the Hadza is rich in fiber, containing 75–100 grams of fiber per day mostly from pulp and seeds of baobab fruits [45]. Their microbiome features reflected the diet, lifestyle, and environment of the study participants, that is, the unindustrialized and traditional rural lifestyle and foraging diet of the hunter-gatherers versus the industrialized and westernized diet and lifestyle of urban Italians. Taken together, there is strong evidence that the gut microbiota metabolizes the plant- and animal-based diet into products that may, respectively, protect against and promote westernized diseases in African populations [42], and will shift compositionally and functionally in favor of the diet and environment at hand [43], thus affecting health.

Nutrition and the Gut Microbiome in the USA and Canada

Nutrition can impact the general well-being of people of every age, race, and ethnicity. Notably, industrialized nations bear witness to an obesity epidemic, setting the stage for vast increases of serious chronic illnesses, such as nonalcoholic fatty liver disease leading to liver failure and metabolic syndrome, which predisposes individuals to diabetes and its multiple micro- and macro-vascular complications. While lifestyle changes through diet and exercise are encouraged to combat the metabolic syndrome, several researchers in Canada and the USA have focused on modulation of the gut microbiota as a viable strategy to manage weight and improve metabolic health. These studies are described below and summarized in Table 2.

Prebiotics/Probiotics

Prebiotics, nondigestible fermentable oligo- and polysaccharides that act as a food source for probiotics (living microbes) were used as possible means of modulating the gut microbiota to manage obesity and improve metabolic health. Three studies investigated the effects of prebiotics administered in different formulations on body composition, appetite control, metabolic markers, and gut microbiota in adults and children. Reimer et al. [46] studied the effect of three kinds of isocaloric snack bars, one containing the prebiotic, 8 grams inulin-type fructans (ITF), one containing whey protein, a control bar, and combination of ITF + whey protein bars administered twice daily for 12 weeks in 96 overweight or obese adults. In a separate study, a daily dose of prebiotic oligofructose-enriched inulin (OI) or an isocaloric amount of control maltodextrin as powder dissolved in

Table 2 Studies of the gut microbiome in USA and Canada from 2015 to 2019

Location of study and reference	Disease or condition	Age of participants	Number and/or nutritional status of participants	Study design	Study outcomes
Chicago, USA Krumbeck et al. 2018	Obesity	18–65 years	$n=94$, 6 treatment groups placebo, probiotic, and prebiotic combinations	Randomized, double-blind, placebo-controlled	Gut microbiota
Toronto, Canada DaSilva et al. 2018	Nonalcoholic fatty liver disease	21–68 years	$n=39$ with biopsy-verified NAFLD, $n=28$ normal controls	Cross-sectional study	Gut microbiota, glucose, HbA1c, lipid profile
Calgary, Canada Reimer et al. 2017	Obesity	18–75 years	$n=125$; $n=27$ control, $n=26$ inulin, $n=21$ whey, $n=22$ inulin + whey	Single-center, placebo-controlled, double-blind study	Gut microbiota
Calgary, Canada Nicolucci et al. 2017	Obesity	7–12 years; male and female	$n=38$; $n=20$ oligofructose-enriched inulin, $n=18$ placebo maltodextrin	Randomized, controlled study	Gut microbiota
Vancouver, Edmonton, Winnipeg/ Winkler-Morden, and Toronto, Canada Stearns et al. 2017	Normal health	1 year	$n=173$ white Caucasian; $n=182$ South Asian	Sub-study	Gut microbiota, fecal fat, short-chain fatty acids
Montreal, Canada Dubois et al. 2017	Normal health	24–67 years (Inuit) 23–48 years (non-Inuit)	$n=15$ Inuit participants; $n=9$ non-Inuit participants	Observational study	Gut microbiota
Canada/USA Maldonado-Gómez et al. 2016	Normal health	22 and 38 years	$n=23$ placebo or AH1206	Double-blind, placebo-controlled, human crossover study	Gut microbiota
Calgary, Canada Lambert et al. 2015	Obesity	44 ± 15 years	$n=50$)9 male, 41 female); placebo or pea fiber	Randomized controlled feeding trial	Gut microbiota, body composition, metabolic markers of obesity
Florida, USA Hanifi et al. 2015	Normal health	18–50 years	$n=81$; 4 treatments, placebo, B. subtilis doses	Double-blind, placebo-controlled randomized trial	Gut microbiota
USA/Canada Krumbeck et al. 2015	Nonalcoholic fatty liver disease model	4-week-old Sprague–Dawley rats	$N=3$–6 rats per 5 treatments groups	Bifidobacteria isolated from human stool	Gut microbiota
Toronto, Canada Rahat-Rozenbloom et al. 2014	Obesity	35.8 ± 4.2 (lean) 42.5 ± 3.9 (overweight and obese)	$n=22$; 11 lean and 11 overweight and obese	Observational study	Rate SCFA absorption, gut microbiota, dietary intake

water was consumed prior to the evening meal for 16 weeks in 38 overweight and obese but otherwise healthy children [47]. ITF, whey protein, and the combination snack bars all improved long-term appetite control but only whey protein was associated with modest reduced body fat in the adult participants. There was no significant additive effect in weight loss or appetite regulation when ITF and whey protein were administered in combination. A significant increase in *Bifidobacterium* was observed only with the ITF-containing snack bars. Among children who consumed the OI prebiotic, researchers observed normalized weight gain, reduced whole body and trunk body fat, and decreased primary fecal bile acids, cholic acid and chenodeoxycholic acid compared to placebo. However, there was no reduction in absolute body weight. *Bifidobacterium* also increased with OI intake compared to placebo. A criticism of the study in children is the distribution of race of the participants was 82% Caucasian and 18% African American and Hispanic. In the third prebiotic study, the effect of a daily dose of 15 grams of yellow pea fiber consumed prior to large meals for 12 weeks in overweight/obese adults with stable weight was examined [48]. The consumption of the yellow pea fiber resulted in small improvements in body fat and glucose tolerance along with a reduction in body weight primarily as fat mass but no significant changes in microbiota were detected at this dose.

Probiotics have been used to increase the proportion of intestinal microbes known to have beneficial effects on human gut mucosa. However, due to their short colonization time, their beneficial effects are limited. In an attempt to produce a probiotic with long-term sustainability, Maldonado-Gómez et al. [49] investigated orally administered *Bifidobacterium longum*. In this double-blind, placebo-controlled, human crossover study, 23 adults consumed a daily dose of 10^{10} viable cells of *B. longum* AH1206 or maltodextrin placebo in random succession with 1 to 3 weeks washout between 2-week treatments and 4-week test of persistence. In 30% of participants, the strain remained detectable in feces for at least 6 months without causing gastrointestinal symptoms or impacting the resident gut microbiota. The long-term persistence of AH1206 was dependent on individualized features of the resident gut microbiota. These results suggest that bacterial species can be established and sustained in the gut of certain individuals, given the opportunity and suitable environment. Krumbeck et al. [50, 51] described testing of a probiotic preparation involving an in vivo selection (IVS) method compared with commercially prepared probiotic administered with (synbiotic) and without a prebiotic. In the first study, researchers isolated a bacterial strain of *Bifidobacterium* (IVS-1) in fecal samples following 9-week administration of increasing doses of prebiotic galactooligosaccharides (GOS) to volunteers. A retrospective analysis of fecal microbiota in one participant demonstrated eightfold enrichment in *Bifidobacterium* post intake. The autochthonist (bacteria already resident in the host) IVS-1 functionality was tested in rats administered the bacterial strain alone, the GOS prebiotic alone, or the combination of the pre- and probiotics. IVS-1 outperformed the resident *Bifidobacterium* in terms of replication in the presence of GOS. Investigators concluded that IVS can be used to successfully formulate a synergistic synbiotic that will overtake other strains naturally resident in the gut [50]. In the second study, researchers used a randomized, double-blind, placebo-controlled, parallel-arm clinical trial to evaluate IVS bifidobacterial strains and commercial bifidobacterial probiotic preparations with and without GOS prebiotic versus placebo in improving intestinal permeability among 94 obese adults. Improvements in intestinal permeability were shown with both probiotic treatments and GOS. The treatment effects were primarily directed to permeability of the colon. This six-arm study did not demonstrate a synergic effect in the two products containing both pre- and probiotics. However, authors stated that the IVS probiotic preparation was more effective than the commercial probiotic in establishing residence in the human gut [51]. The latter two studies represent research examining autochthonist versus commercially prepared probiotics and exploring potential synergism with specific prebiotics. Clearly, these investigators promote the use of individually isolated probiotic strains naturally resident in the gut over the introduction of foreign strains that are commercially prepared. This practice would be closely aligned with holistic approaches to health care and traditional indigenous healing practices.

Traditional Foods and Ethnicity

While host genetics and lifestyle influence the gut microbiome, we know that diet is the major factor shaping it. Investigators in this study were interested in how seasonality affects the gut microbiome among Canadian Inuit people, assuming that variations in food sources available during different seasons would contribute to large shifts in gut microbiome composition [52].

Stool samples collected from toilet tissue were analyzed for microbiome composition among Canadian Inuit volunteers living in Resolute Bay, Nunavut, and Canadian individuals of primarily European descent living in Montreal, Quebec. Important findings were that 45–61% (the greatest proportion) of variability in microbiome composition was due to individual identity. Examining both groups together, 11% of the variability over the course of 12 months was explained by diet. Among Inuit alone, 17% of the variability was due to diet. Sex explained 3–4% of the variability in microbiomes, and geography contributed 3–5% to the variability. Microbiomes of women varied more than men, and microbiomes among Nunavut residents were more varied

than Montreal residents. Unexpectedly, there were no significant seasonal shifts in microbiome composition in either the Inuit in Nunavut or the European descent Montreal groups. Aside from the above results, the study revealed that Canada's indigenous population living in its northern remote areas is no longer solely reliant on "traditional" food items. Westernized foods are available year-around, which could be the reason why seasonality did not seem to affect microbiome composition in this study. However, the Inuit microbiome did vary over time more than the Montreal diet. Authors suspect greater food security in Montreal and the fact that all foods are available year-round in that area were the reasons for less variability over time in that group. Circumstances in Nunavut resemble those of other indigenous communities in the circumpolar north. Elders and other community residents report less reliance on subsistence lifestyle with the advent of big box stores (e.g., Costco, Walmart, Sam's Club) in more urbanized areas, increased mechanization and commercial air transport, Internet orders through companies that provide free or substantially reduced shipping fees, and the availability of "westernized" prepared foods in the local community stores. A second important study outcome was the assessment of dietary intake measured as mean frequency of consumption (number of times eaten per participant) of each food category per day. The Nunavut group consumed substantially more eggs (mean frequency 0.91 vs. 0.18) than the Montreal group, while the Montreal group consumed more dairy products (mean frequency 1.9 vs. 0.88) and alcoholic beverages (mean frequency 0.96 vs. 0.22). There was also notably less consumption of fruits and vegetables (mean frequency 0.85 vs. 2.72) among the Nunavut group than the Montreal group. Importantly, the Nunavut group diet consisted largely of game meats—raw, cooked, and fermented. The foods traditionally fermented include walrus and whale meat, fish, seal, and caribou. The meat and fat that is caught in the summer is buried in the ground over the autumn and winter months ready for consumption in the next year. These traditional foods are similar to traditional foods consumed by indigenous peoples (Inupiat and Yup'ik) in northern circumpolar regions of Alaska. In Alaska, raw, cooked, and fermented game sources include moose, caribou, and marine mammals, such as whale meat and blubber, sea lion, walrus, seal and seal oil, wild birds and bird eggs, and multiple ocean and river fish. However, all communities have local markets, which stock "western" prepared/preserved foods with long shelf life, decreasing reliance on traditional indigenous food sources.

The developing gastrointestinal microbiota in the first years of life is important for immune function, nutrient metabolism, and protection from pathogens [53, 54]. Early evidence suggests that diet influences the infant gut microbiome [55]. There is preliminary evidence that gut microbial composition in adults and children varies by age, dietary intake, ethnicity, geography, and adoption of western lifestyles [56, 57]. In 2017, Stearns et al. [58] explored the effects of race/ethnicity separate from geographic region on the gut microbiota in early life. Participants from two prospective Canadian birth cohorts were included. Researchers used 1-year fecal samples from 173 Caucasian infants and 182 infants of South Asian ancestry living in Canada in the main analysis. Results demonstrated that the gut microbiome of infants is influenced by race/ethnicity, age, weight gain, and breast-feeding. South Asian mothers lived in Canada for an average of 8 years (vs. a lifetime for Caucasian mothers), were younger, more likely to be vegetarian, and more likely to be diagnosed with gestational diabetes than the Caucasian mothers. South Asian infants had higher abundance of several members of lactic acid bacteria (LAB) contained in the feces, specifically *Bifidobacterium*, *Lactococcus*, *Streptococcus*, and *Enterococcus* than in the Caucasian infants feces. LAB are responsible for the breakdown of carbohydrates to lactate and acetate [57, 59]. The abundance of the *Atopobium* cluster of *Actinobacteria* was also higher in South Asian vs. Caucasian infants. This group of bacteria is saccharolytic and has been observed in abundance in the microbiome of individuals with a fiber-rich diet [60]. Caucasian infants showed greater abundance of members of *Firmicutes* from the order *Clostridiales*, shown to increase in response to diets rich in animal protein and fat [61]. This study demonstrates race/ethnicity and infant feeding practices play important roles in the development of the infant gut microbiome. While race/ethnicity is not modifiable, the infant diet is and its alteration can assist in promoting healthy gut microbiota.

Nutrition and the Gut Microbiome in Europe

Numerous recent clinical studies performed by research groups in Europe examine the effects of whole foods and additives on the gut microbiome and human health. While European diets are quite diverse, the general trend toward "Westernization" of diet (e.g., high intake of fat and low consumption of milk) causes similar health problems in most European countries (e.g., rising incidences of obesity, cardiovascular diseases, and colon cancer). In this context of nutrition, gut microbiome and human health, a selection of recent studies is discussed (Table 3).

Nuts (walnuts in particular) are known to have health benefits in that their consumption was shown to lower plasma lipid levels [62]. The aim of one study was to investigate how walnut intake alters the gut microbiome composition and diversity. In a randomized controlled trial (crossover design) of 96 healthy participants, a walnut-enriched diet was administered for 8 weeks followed by a switch to nut-free diet. [63]. A second group of 98 participants followed

Table 3 Studies of the gut microbiome in Europe and Asia from 2015 to 2019

Location of study and reference	Disease or condition	Age of participants	Number and/or nutritional status of participants	Study design	Study outcomes
Russia Volokh et al. 2019	Normal health	18–40 years; male and female	$n=150$ fermented dairy product	Part of large open prospective controlled study	Gut microbiota
Germany Bamberger et al. 2018	Normal health	63 ± 7 years; male and female	$n=96$ intervention (walnuts) first; $n=98$ control first	Randomized, controlled, prospective, crossover study	Gut microbiota,
Germany Trautvetter et al. 2018	Normal health	29 ± 7 years; male and female	$n=62$ intervention (phosphorus and calcium); $n=62$ control first	Double-blind randomized controlled intervention trial	Gut microbiota, fecal fat, short-chain fatty acids
Sweden Zhang et al. 2018	Normal health	39 ± 10.1; 34.7 ± 10.4; 33.5 ± 7.4	$n=17$ switched to vegetarian diet $n=14$ control diets (seven omnivorous, seven vegetarian)	Sub-study of a randomized trial	Gut microbiota, immune analysis
China Wan et al. 2019	Normal health	18–35 years	$n=217$; $n=73$ lower-fat, $n=73$ moderate-fat, $n=71$ higher-fat diets	Randomized controlled feeding trial	Gut microbiota, fecal metabolomics, plasma proinflammatory factors
Japan Kushida et al. 2019	Normal health	20–29 years	$n=11$ 1975 Japanese diet, $n=10$ modern Japanese diet	Randomized controlled clinical trial	Gut microbiota
China Zhao et al. 2018	Type 2 diabetes	Adults	$n=43$ type 2 diabetes; $n=16$ control diet or $n=27$ WTP diet	Randomized clinical trial	Gut microbiota, hemoglobin A1c (HbA1c)
China Yuan et al. 2018	Normal health or obese	27–46 years	$n=12$, green tea liquid	Observational study	Gut microbiota, oral microbiota
China Zhang et al. 2017	Obesity	3–16 years	$n=17$ Prader–Willi syndrome; $n=21$ simple obesity intervention: diet of whole grains, traditional Chinese medicinal foods, prebiotics (WTP)	Open-labeled and self-controlled clinical trial	Gut microbiota, urinary metabolites, fecal metabolites, inflammation analysis
China Xie et al. 2017	Epilepsy	1.95 ± 3.10 years (epilepsy), up to 3 years (healthy control)	$n=14$ epilepsy, $n=30$ control before and after ketogenic diet	Controlled clinical trial	Gut microbiota
China Li et al. 2017	Constipation	Adults under 65 years	$n=100$; $n=50$ (Deshipu stachyose granules), $n=50$ (control)	Nonblind randomized control trial	Gut microbiota, bowel function
China Xu et al. 2015	Type 2 diabetes	54.23 ± 8.56 (placebo), 55.06 ± 7.49 (low dose), 53.18 ± 8.89 (median dose), 51.24 ± 9.10 (high dose)	$n=187$; $n=44$ high dose, $n=52$ moderate dose, $n=50$ low dose Gegen Qinlian Decoction, $n=41$ placebo	Randomized double-blinded, placebo-controlled clinical trial	Gut microbiota, fasting blood glucose, HbA1c,

the dietary pattern in reverse order, and fecal samples were collected for 16S rRNA gene sequencing analysis. No difference was found in alpha-diversity, but with regard to beta-diversity a distinct clustering of the walnut and control groups was observed. However, the walnut diet explained only about 5% of observed difference compared to the control diet. In the walnut group, a significantly increased abundance of *Ruminococcaceae* and *Bifidobacteria* coincided with a decrease in *Clostridium* sp. Cluster XIVa species compared to control diet. Thus, walnut intake may promote compositional shifts of the gut microbiota to potentially probiotic and SCFA-producing species that may account for health benefits associated with walnut consumption. However, the link to health-promoting, walnut-dependent metabolites remains speculative, since this study reported compositional differences and did not quantify actual microbial metabolites.

A vegetarian diet rich in vitamins and fibers has been shown to be beneficial to the immune system [64]. The aim of another recent study was to investigate the effect of a 3-month lacto-ovo-vegetarian diet on the gut microbiota and the immune system in healthy omnivorous volunteers [65]. Fifteen healthy omnivorous volunteers switched to a lacto-ovo-vegetarian diet for 3 months, and blood and fecal samples were collected pre- (month 0) and post-intervention (month 3). Control groups consisted of healthy volunteers staying on their omnivorous diet or vegetarian diet, respectively. All fecal samples were analyzed by metagenomic shotgun sequencing. While no difference in alpha-diversity was detected, a few genus-level changes were associated with the vegetarian diet intervention. After 3 months of lacto-ovo-vegetarian diet, the abundance of *Alistipes* was reduced, coinciding with increased abundance of *Roseburia inunilivorans*, *Ruminococcus lactis*, *Lactobacillus plantarum*, and *Streptococcus thermophiles* in the fecal microbiota. However, it needs to be noted that the authors did neither comment on compliance of study participants that switched from an omnivorous to a vegetarian diet nor provided details on how/whether dietary intake was recorded, which are crucial parameters to evaluate the obtained results. An additional comparison of both control groups (long-term omnivores and vegetarians) revealed compositional differences at genus and species levels, supporting the idea that long-term dietary patterns are a major driver of gut microbiota assembly. In conclusion, a switch to the vegetarian diet had an impact on gut microbiota composition, but its functional relevance on gut microbial co-metabolism remains to be elucidated.

It is reported that *Bifidobacteria* can have functional benefits by cross-feeding other members of gut microbiota specialized on production of butyrate, an essential short-chain fatty acid for colon epithelial cells [66]. The aim of this recent study was to analyze how consumption of fermented dairy products (FDP)—a yogurt fortified with the probiotic *Bifidobacterium animalis subsp. lactis*, BB-12, may be associated with positive impact on human health [67]. In this prospective intervention study, 150 healthy participants received FDP fortified with BB-12 twice a day for 30 days in total. Fecal samples were collected at days 0 and 30 and 16S rRNA gene sequencing analysis performed. While no change in alpha-diversity after FDP intake was shown, a correlation analysis of species occurring together identified five major microbial networks and distinct compositional shifts following FDP intake were observed. A paired comparison revealed that abundance of 39 taxa increased and 24 taxa decreased after FDP intake. An increased abundance after FDP intake was demonstrated for *Coriobacteriaceae* (ability to metabolize lactose to lactate), *Bifidobacteriaceae* (probiotic, anti-inflammatory), *Staphylococcaceae*, and *Erysipelotrichaceae*. Decreased numbers were demonstrated for *Lachnoclostridium*_unclassified (involved in bile acid metabolism), and *Roseburia* genera (potential SCFA producers), for example. When investigating changes in lactose-fermenting taxa (e.g., *Bifidobacterium*, *Lactobacillus*, *Lactococcus*), which are likely to be affected by altered FDP intake, the authors identified two clusters of study participants: Cluster 1 showed a weaker increase in relative abundance of lactose-fermenting bacteria compared to the individuals from cluster 2 and less changes in overall fecal microbiota composition. In contrast, a higher number of taxa in the fecal microbiota of individuals from cluster 2 were affected by FDP intake, suggesting that cluster 2 can be termed "responders." Interestingly, these responders showed a lower abundance of lactose-fermenting bacteria in the fecal microbiota at baseline compared to cluster 1 (or "nonresponders"), which may explain the observed cluster-specific increase in lactose-fermenting bacteria. Overall, significant shifts in gut microbiota composition suggest changes induced by 30 days of FDP intake, but the effect on microbial metabolism, in particular with regard to human health, remains unclear.

According to the National Health and Nutrition Examination Survey of the USA, dietary phosphate intake of Americans exceeds the daily recommended intake of 700 mg phosphorus for adults and the elderly [68]. This is mainly a result of the increased consumption of food prepared or treated with phosphate additives, ranging from baked goods to cola beverages [69]. A recent study by Trautvetter et al. [70] aimed at analyzing the impact of high phosphorus intake on gut-related parameters. The randomized controlled trial, double-blinded intervention (monosodium phosphate with/without calcium carbonate supplements) with 62 healthy participants consisted of a pre-intervention phase (consumption of placebo for 2 weeks) and an intervention for 8 weeks (monosodium phosphate with or without calcium carbonate). Fecal samples were collected after placebo and

intervention, and 16S rRNA gene sequencing, fecal water cytotoxicity, and targeted short-chain fatty acids were analyzed. Significantly higher levels of total short-chain fatty acids and acetate in feces were detected after 8 weeks on monosodium phosphate with calcium carbonate compared to monosodium phosphate without calcium carbonate. However, no differences in epithelial cell cytotoxicity and only minor differences in gut microbiota composition between men from both groups, but not women were found (e.g., higher abundance of *Clostridium* XVIII in men receiving the highest levels of monosodium phosphate and calcium carbonate compared to the other groups). These results suggest that there is no strong effect of a high phosphorus diet on gut microbiota composition and metabolism, despite small increases in short-chain fatty acid production.

Nutrition and the Gut Microbiome in Asia

Weight management, obesity, and metabolic disorders are also a concern in Asian populations as diet and lifestyles change. The following Asian studies break down the effects of food components and different diets on the gut microbiome and how they may influence health (summarized in Table 3).

Fat

In China, the nutritional transition from a traditional low-fat, high-carbohydrate diet to a diet relatively higher in fat and lower in carbohydrate has been associated with a dramatic increase in the risk of obesity, type 2 diabetes, cardiovascular diseases, and colon cancer in the past 30 years [71]. Studies have shown that human gut microbiota diversity and richness are reduced when comparing high-fat diets with more traditional diets of relatively higher proportions of carbohydrates [72, 73]. Wan et al. [74] performed a 6-month randomized controlled feeding trial of 217 healthy young adults, varying the amounts of fat in isocaloric diets containing lower fat (20% energy), moderate fat (30% energy), and higher fat (40% energy). At the phylum level, the moderate- and higher-fat diets decreased the ratio of *Firmicutes* to *Bacteroidetes* after intervention. At the genus level, the higher-fat diet decreased the abundance of *Faecalibacterium*, increased the abundance of *Alistipes* and *Bacteroides*, while the lower-fat diet increased the *Faecalibacterium* and *Blautia* abundance after the intervention. Concentration of fecal butyrate was increased after the lower-fat diet intervention and decreased after the higher-fat intervention. The changes in relative abundance of *Blautia* were negatively associated with the changes in serum total cholesterol, low-density lipoprotein cholesterol, and nonhigh-density lipoprotein cholesterol, whereas the change in *Bacteroides* abundance was positively correlated with the changes in these blood lipid markers. Higher-fat consumption by healthy young adults whose diet is in a state of nutrition transition appeared to be associated with unfavorable changes in gut microbiota, fecal metabolomic profiles, and plasma proinflammatory factors, which might confer adverse consequences for long-term health outcomes [74]. It should, however, be noted that these diets were kept isocaloric, so it would be fair to replace low fat with high carbohydrate, and high fat with low carbohydrate. Further, the low-carbohydrate diet would have lower fiber. In this context, international studies that reflect different environmental conditions are important to identify the effects of diet composition on the gut microbiota. For example, Ocvirk et al. recently showed that the high-fat, low-carbohydrate, and fiber diet of the Alaska Native people (AN) was associated with a very different colonic microbiota composition. In this study, the fecal microbiota showed a compositional predominance of *Blautia* and fecal levels of short-chain fatty acids were significantly lower compared to a low-risk rural African population. The importance of this is that AN have the highest documented risk of colon cancer in the world, and the incidence of other westernized diseases such as obesity, diabetes, and cardiovascular diseases is also high. This highlights the concern about conclusions that dysbiotic signatures may play a causative role in westernized diseases [75].

Fiber

The need to determine whether associations between diet, the microbiome, and westernized diseases were indeed causal led to very interesting human trial-experimental animal mechanistic studies reported by Zhao et al. in Science. In a randomized clinical study of Chinese patients with type 2 diabetes mellitus, 43 patients received either the usual care based on the 2013 recommendations of the Chinese Diabetes Society (control) or a high-fiber diet (approximately 50 grams/day) composed of whole grains, traditional Chinese medicinal foods, and prebiotics (WTP) for 84 days [76]. Daily energy and macronutrients were similar across groups. Both groups received acarbose as the standardized medication (transforms part of the starch in the diet into a fiber by reducing its digestion and making it more available as fermentable carbohydrate in the colon). A group of 15 SCFA-producing strains that were selectively promoted by the WTP diet were detected (Fig. 2) [76]. Promoting this active group of SCFA producers not only enhanced a beneficial function but also maintained a gut environment that keeps detrimental bacteria such as indole- and hydrogen sulfide-producing strains at bay. Hemoglobin A1c (HbA1c) levels (primary

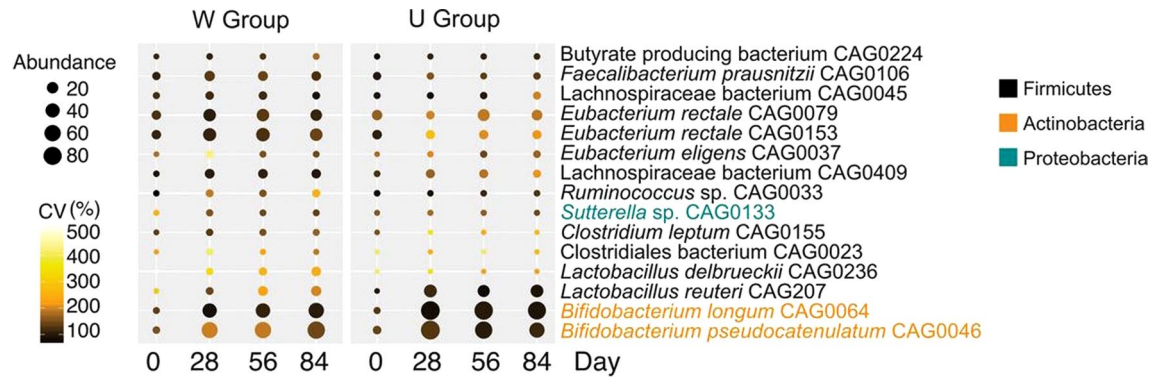


Fig. 2 From the study of Zhao et al., 43 patients with T2DM were randomized for 84 days to a traditional Chinese high-fiber diet. Eighty nine percent of the high-fiber group showed normalization of their HbA1c compared to 29% of the control group. Shotgun sequencing identified 15 specific SCFA-producing strains (see above) that were promoted by dietary fiber which were also associated with better improvement in hemoglobin A1c levels and diminished indole and hydrogen sulfide producers [76]

outcome measure in this study) were significantly decreased in both groups from baseline in a time-dependent manner, but by day 28 of the study the decrease was greater in the WTP group, partly via increased glucagon-like peptide-1. There was a temporal difference in fasting blood glucose levels—only the WTP group achieved a significant reduction by day 28, although at the end of the intervention there was no difference between groups. The WTP group also showed greater reduction in body weight and better blood lipid profiles than the control group. In another Chinese study, Zhang et al. performed a hospitalized dietary interventional trial for 30 days of 40 children diagnosed with genetic (diagnosed with Prader–Willi syndrome (PWS)) and simple obesity. Some of the children in the PWS group continued on the diet intervention for 60 days. A WTP diet in combination with appropriate amounts of vegetables, fruits, and nuts induced significant weight loss and concomitant structural changes of the gut microbiota together with reduction in serum antigen load and alleviation of inflammation [77]. This dietary intervention shifted the dysbiotic gut microbiota to a healthier structure with relatively lower levels of bacteria that can produce potentially toxic metabolites from the fermentation of dietary fats and proteins, and higher level of bacteria that can produce potentially beneficial products from the fermentation of carbohydrates.

Chinese Traditional Herbal Formulas

A traditional Chinese herbal formula, Gegen Qinlian Decoction (GQD), is believed to alleviate type 2 diabetes. A 12-week randomized double-blinded placebo-controlled clinical trial in 187 patients with type 2 diabetes by Xu et al. [78] found that low (48 grams), moderate (144 grams), and high doses (240 grams) of GQD induced structural changes of the gut microbiota composition and enriched the amounts of "beneficial" bacteria, such as *Faecalibacterium* spp., high butyrate producers, in a dose-dependent manner. Another herbal formulation, the prebiotic Deshipu stachyose granules (DSG), a mixture of α-galactooligosaccharides derived from the dietary roots of Lycopus lucidus Turcz, was reported to facilitate intestinal peristalsis and defecation relieving constipation in mice [79]. A study designed to investigate the effect of DSG on gut microbiota and bowel function in 100 adult volunteers found that DSG at a dosage of 5 grams per day for 14 days could instructively regulate the gut microbiota with significant increases in "probiotic" species *bifidobacteria* and *lactobacilli* and a remarkable decrease in the potential pathobiont *Clostridium perfringens*. DSG also could effectively improve the bowel function of patients suffering from constipation with a marked increase in defecation frequency from 1.78 times per week to 3.02 times per week [80].

Green tea has been claimed to hold health benefits including anti-colorectal cancer activity and is characterized by the presence of large amounts of polyphenols, also known as catechins, such as epigallocatechin-3-gallate, epigallocatechin, epicatechin gallate, and epicatechin [81]. Yuan et al. [82] investigated the effect of green tea liquid (GTL) consumption (400 ml per day for 2 weeks) on the gut and oral microbiome in 12 healthy volunteers. They found an increased FIR:BAC (Firmicutes-to-Bacteroidetes ratio), elevated SCFA-producing genera, and reduction in bacterial LPS synthesis in feces following a 1-week washout period. GTL also altered the salivary microbiota and reduced the functional pathways abundance relevant to carcinogenesis and reduced the fecal levels of Fusobacterium.

Ketogenic Diets

A ketogenic diet (KD) consists of a diet very low in carbohydrates and high in fat. This reduction in carbohydrates puts the body in a state of ketosis due to the need to increase the mobilization and oxidation of fat for energy. A KD has been advocated for the treatment for chronic epilepsy. Xie et al. [83] performed a comparison between 14 infants with epilepsy and 30 healthy control infants before and after KD treatment to explore whether the gut microbiota of infants with refractory epilepsy differed with that of age-matched healthy infants. The therapeutic effect of the KD on refractory epilepsy and the changes in the gut microbiota were also studied. After being on the KD treatment for a week, 64% of epileptic infants showed an improvement with a 50% decrease in seizure frequency. The gut microbiome structure in epileptic infants differed dramatically from that in healthy infants. Proteobacteria, which were significantly higher in epileptic infants, decreased dramatically after the KD. *Cronobacter* predominated in the epileptic infants group and remained at a low level both in the healthy infants and in the epileptic infants after the KD. *Bacteroides* increased significantly in the epileptic infants after KD, in which *Prevotella* and *Bifidobacterium* also grew in numbers and kept increasing [83].

Summary

The studies discussed provide irrefutable evidence from around the world that the human microbiome can be modified by dietary change in children and adults. While the associations between specific dietary components and health and disease are strong, a lot more work on causality is needed before specific microbes, or groups of microbes, can be used therapeutically. The evidence for a balanced diet and balanced microbiota being able to prevent westernized diseases and extend good-quality life span is, however, compelling [11].

Compliance with Ethical Standards

Conflict of interest ASW, CC, and SJO report research support from the NIH. KRK, CAF, and ZTM report research support from the NIH and Mayo Clinic outside of this work. FRS reports research support from the NIH outside of this work. All other authors have no conflict of interest to disclose.

References

1. Sender R, Fuchs S, Milo R. Revised estimates for the number of human and bacteria cells in the body. *PLoS Biol*. 2016;14(8):e1002533.
2. Subramanian S, Huq S, Yatsunenko T, et al. Persistent gut microbiota immaturity in malnourished Bangladeshi children. *Nature*. 2014;510(7505):417–421.
3. Arnold JW, Roach J, Azcarate-Peril MA. Emerging technologies for gut microbiome research. *Trends Microbiol*. 2016;24(11):887–901.
4. Reid G, Nduti N, Sybesma W, et al. Harnessing microbiome and probiotic research in sub-Saharan Africa: recommendations from an African workshop. *Microbiome*. 2014;16(2):12.
5. Graf D, Di Cagno R, Fåk F, et al. Contribution of diet to the composition of the human gut microbiota. *Microb Ecol Health Dis*. 2015;4(26):26164.
6. Walker AW, Ince J, Duncan SH, et al. Dominant and diet-responsive groups of bacteria within the human colonic microbiota. *ISME J*. 2011;5(2):220–230.
7. Bäckhed F, Ley RE, Sonnenburg JL, Peterson DA, Gordon JI. Host-bacterial mutualism in the human intestine. *Science*. 2005;307(5717):1915–1920.
8. Topping DL, Clifton PM. Short-chain fatty acids and human colonic function: roles of resistant starch and nonstarch polysaccharides. *Physiol Rev*. 2001;81(3):1031–1064.
9. O'Keefe SJD. Diet, microorganisms and their metabolites, and colon cancer. *Nat Rev Gastroenterol Hepatol*. 2016;13(12):691–706.
10. O'Keefe SJ. The association between dietary fibre deficiency and high-income lifestyle-associated diseases: Burkitt's hypothesis revisited. *Lancet Gastroenterol Hepatol*. 2019;4(12):984–996.
11. O'Keefe SJD. Plant-based foods and the microbiome in the preservation of health and prevention of disease. *Am J Clin Nutr*. 2019;110:265–266.
12. Kim Y, Je Y. Dietary fibre intake and mortality from cardiovascular disease and all cancers: a meta-analysis of prospective cohort studies. *Arch Cardiovasc Dis*. 2016;109(1):39–54.
13. Park Y, Subar AF, Hollenbeck A, Schatzkin A. Dietary fiber intake and mortality in the NIH-AARP diet and health study. *Arch Intern Med*. 2011;171(12):1061–1068.
14. Aune D, Chan DSM, Lau R, et al. Dietary fibre, whole grains, and risk of colorectal cancer: systematic review and dose-response meta-analysis of prospective studies. *BMJ*. 2011;10(343):d6617.
15. Blanton LV, Barratt MJ, Charbonneau MR, Ahmed T, Gordon JI. Childhood undernutrition, the gut microbiota, and microbiota-directed therapeutics. *Science*. 2016;352(6293):1533.
16. Black RE, Victora CG, Walker SP, et al. Maternal and child undernutrition and overweight in low-income and middle-income countries. *Lancet*. 2013;382(9890):427–451.
17. Nel E. Severe acute malnutrition. *Curr Opin Clin Nutr Metab Care*. 2018;21(3):195–199.
18. Jones KD, Thitiri J, Ngari M, Berkley JA. Childhood malnutrition: toward an understanding of infections, inflammation, and antimicrobials. *Food Nutr Bull*. 2014;35(2 Suppl):S64–S70.
19. Kau AL, Ahern PP, Griffin NW, Goodman AL, Gordon JI. Human nutrition, the gut microbiome and the immune system. *Nature*. 2011;474(7351):327–336.
20. Smith MI, Yatsunenko T, Manary MJ, et al. Gut microbiomes of Malawian twin pairs discordant for kwashiorkor. *Science*. 2013;339(6119):548–554.
21. Oldenburg CE, Sié A, Coulibaly B, et al. Effect of commonly used pediatric antibiotics on gut microbial diversity in preschool

children in burkina faso: a randomized clinical trial. *Open Forum Infect Dis.* 2018;5(11):ofy289.
22. Atukunda P, Muhoozi GKM, van den Broek TJ, et al. Child development, growth and microbiota: follow-up of a randomized education trial in Uganda. *J Glob Health*. 2019;9(1):010431.
23. Paganini D, Uyoga MA, Kortman GAM, et al. Iron-containing micronutrient powders modify the effect of oral antibiotics on the infant gut microbiome and increase post-antibiotic diarrhoea risk: a controlled study in Kenya. *Gut*. 2019;68(4):645–653.
24. Jaeggi T, Kortman GAM, Moretti D, et al. Iron fortification adversely affects the gut microbiome, increases pathogen abundance and induces intestinal inflammation in Kenyan infants. *Gut.* 2015;64(5):731–742.
25. Tang M, Frank DN, Hendricks AE, et al. Iron in micronutrient powder promotes an unfavorable gut microbiota in Kenyan infants. *Nutrients*. 2017;9(7):776.
26. Davis JCC, Lewis ZT, Krishnan S, et al. Growth and morbidity of Gambian infants are influenced by maternal milk oligosaccharides and infant gut microbiota. *Sci Rep*. 2017;12(7):40466.
27. Ordiz MI, Stephenson K, Agapova S, et al. Environmental enteric dysfunction and the fecal microbiota in Malawian children. *Am J Trop Med Hyg*. 2017;96(2):473–476.
28. Million M, Tidjani Alou M, Khelaifia S, et al. Increased gut redox and depletion of anaerobic and methanogenic prokaryotes in severe acute malnutrition. *Sci Rep*. 2016;17(6):26051.
29. Charbonneau MR, O'Donnell D, Blanton LV, et al. Sialylated milk oligosaccharides promote microbiota-dependent growth in models of infant undernutrition. *Cell*. 2016;164(5):859–871.
30. Kristensen KHS, Wiese M, Rytter MJH, et al. Gut microbiota in children hospitalized with oedematous and non-oedematous severe acute malnutrition in Uganda. *PLoS Negl Trop Dis*. 2016;10(1):e0004369.
31. Reyes A, Blanton LV, Cao S, et al. Gut DNA viromes of Malawian twins discordant for severe acute malnutrition. *Proc Natl Acad Sci USA*. 2015;112(38):11941–11946.
32. Kau AL, Planer JD, Liu J, et al. Functional characterization of IgA-targeted bacterial taxa from undernourished Malawian children that produce diet-dependent enteropathy. *Sci Transl Med*. 2015;7(276):276ra24.
33. Bwakura-Dangarembizi M, Amadi B, Bourke CD, et al. Health outcomes, pathogenesis and epidemiology of severe acute malnutrition (HOPE-SAM): rationale and methods of a longitudinal observational study. *BMJ Open*. 2019;9(1):e023077.
34. Vray M, Hedible BG, Adam P, et al. A multicenter, randomized controlled comparison of three renutrition strategies for the management of moderate acute malnutrition among children aged from 6 to 24 months (the MALINEA project). *Trials*. 2018;19(1):666.
35. Kane AV, Dinh DM, Ward HD. Childhood malnutrition and the intestinal microbiome. *Pediatr Res*. 2015;77(1–2):256–262.
36. Gordon JI, Dewey KG, Mills DA, Medzhitov RM. The human gut microbiota and undernutrition. *Sci Transl Med*. 2012;4(137):137ps12.
37. Williams JE, Price WJ, Shafii B, et al. Relationships among microbial communities, maternal cells, oligosaccharides, and macronutrients in human milk. *J Hum Lact*. 2017;33(3):540–551.
38. Gough EK, Stephens DA, Moodie EEM, et al. Linear growth faltering in infants is associated with Acidaminococcus sp. and community-level changes in the gut microbiota. *Microbiome*. 2015;3:24.
39. Sonnenburg ED, Sonnenburg JL. Starving our microbial self: the deleterious consequences of a diet deficient in microbiota-accessible carbohydrates. *Cell Metab*. 2014;20(5):779–786.
40. Ferlay J, Shin H-R, Bray F, Forman D, Mathers C, Parkin DM. Estimates of worldwide burden of cancer in 2008: GLOBOCAN 2008. *Int J Cancer*. 2010;127(12):2893–2917.
41. Katsidzira L, Ocvirk S, Wilson A, et al. Differences in fecal gut microbiota, short-chain fatty acids and bile acids link colorectal cancer risk to dietary changes associated with urbanization among Zimbabweans. *Nutr Cancer*. 2019;71(8):1313–1324.
42. O'Keefe SJD, Li JV, Lahti L, et al. Fat, fibre and cancer risk in African Americans and rural Africans. *Nat Commun*. 2015;28(6):6342.
43. Rampelli S, Schnorr SL, Consolandi C, et al. Metagenome sequencing of the Hadza A. *Curr Biol*. 2015;25(13):1682–1693.
44. Turroni S, Fiori J, Rampelli S, et al. Fecal metabolome of the Hadza hunter-gatherers: a host-microbiome integrative view. *Sci Rep*. 2016;14(6):32826.
45. de Vrieze J. Gut instinct. *Science*. 2014;343(6168):241–243.
46. Reimer RA, Willis HJ, Tunnicliffe JM, Park H, Madsen KL, Soto-Vaca A. Inulin-type fructans and whey protein both modulate appetite but only fructans alter gut microbiota in adults with overweight/obesity: a randomized controlled trial. *Mol Nutr Food Res*. 2017;61(11):1700484.
47. Nicolucci AC, Hume MP, Martínez I, Mayengbam S, Walter J, Reimer RA. Prebiotics reduce body fat and alter intestinal microbiota in children who are overweight or with obesity. *Gastroenterology*. 2017;153(3):711–722.
48. Lambert JE, Parnell JA, Tunnicliffe JM, Han J, Sturzenegger T, Reimer RA. Consuming yellow pea fiber reduces voluntary energy intake and body fat in overweight/obese adults in a 12-week randomized controlled trial. *Clin Nutr*. 2017;36(1):126–133.
49. Maldonado-Gómez MX, Martínez I, Bottacini F, et al. Stable engraftment of bifidobacterium longum AH1206 in the human gut depends on individualized features of the resident microbiome. *Cell Host Microbe*. 2016;20(4):515–526.
50. Krumbeck JA, Maldonado-Gomez MX, Martínez I, et al. In vivo selection to identify bacterial strains with enhanced ecological performance in synbiotic applications. *Appl Environ Microbiol*. 2015;81(7):2455–2465.
51. Krumbeck JA, Rasmussen HE, Hutkins RW, et al. Probiotic bifidobacterium strains and galactooligosaccharides improve intestinal barrier function in obese adults but show no synergism when used together as synbiotics. *Microbiome*. 2018;6(1):121.
52. Dubois G, Girard C, Lapointe F-J, Shapiro BJ. The inuit gut microbiome is dynamic over time and shaped by traditional foods. *Microbiome*. 2017;5(1):151.
53. Falk PG, Hooper LV, Midtvedt T, Gordon JI. Creating and maintaining the gastrointestinal ecosystem: what we know and need to know from gnotobiology. *Microbiol Mol Biol Rev*. 1998;62(4):1157–1170.
54. Guarner F, Malagelada J-R. Gut flora in health and disease. *Lancet*. 2003;361(9356):512–519.
55. Bokulich NA, Chung J, Battaglia T, et al. Antibiotics, birth mode, and diet shape microbiome maturation during early life. *Sci Transl Med*. 2016;8(343):343ra82.
56. Yatsunenko T, Rey FE, Manary MJ, et al. Human gut microbiome viewed across age and geography. *Nature*. 2012;486(7402):222–227.
57. Schnorr SL. The diverse microbiome of the hunter-gatherer. *Nature*. 2015;518(7540):S14–S15.
58. Stearns JC, Zulyniak MA, de Souza RJ, et al. Ethnic and diet-related differences in the healthy infant microbiome. *Genome Med*. 2017;9(1):32.
59. Duncan SH, Louis P, Flint HJ. Lactate-utilizing bacteria, isolated from human feces, that produce butyrate as a major fermentation product. *Appl Environ Microbiol*. 2004;70(10):5810–5817.
60. Martínez I, Lattimer JM, Hubach KL, et al. Gut microbiome composition is linked to whole grain-induced immunological improvements. *ISME J*. 2013;7(2):269–280.

61. Wu GD, Chen J, Hoffmann C, et al. Linking long-term dietary patterns with gut microbial enterotypes. *Science*. 2011;334(6052):105–108.
62. Bamberger C, Rossmeier A, Lechner K, et al. A walnut-enriched diet reduces lipids in healthy caucasian subjects, independent of recommended macronutrient replacement and time point of consumption: a prospective, randomized, controlled trial. *Nutrients*. 2017;9(10):1097.
63. Bamberger C, Rossmeier A, Lechner K, et al. A walnut-enriched diet affects gut microbiome in healthy Caucasian subjects: a randomized, controlled trial. *Nutrients*. 2018;10(2):244.
64. Tanaka T, Kouda K, Kotani M, et al. Vegetarian diet ameliorates symptoms of atopic dermatitis through reduction of the number of peripheral eosinophils and of PGE2 synthesis by monocytes. *J Physiol Anthropol Appl Human Sci*. 2001;20(6):353–361.
65. Zhang C, Björkman A, Cai K, et al. Impact of a 3-months vegetarian diet on the gut microbiota and immune repertoire. *Front Immunol*. 2018;27(9):908.
66. Rivière A, Selak M, Lantin D, Leroy F, De Vuyst L. Bifidobacteria and butyrate-producing colon bacteria: importance and strategies for their stimulation in the human gut. *Front Microbiol*. 2016;28(7):979.
67. Volokh O, Klimenko N, Berezhnaya Y, et al. Human gut microbiome response induced by fermented dairy product intake in healthy volunteers. *Nutrients*. 2019;11(3):547.
68. Adatorwovor R, Roggenkamp K, Anderson JJB. Intakes of calcium and phosphorus and calculated calcium-to-phosphorus ratios of older adults: NHANES 2005–2006 data. *Nutrients*. 2015;7(11):9633–9639.
69. Ritz E, Hahn K, Ketteler M, Kuhlmann MK, Mann J. Phosphate additives in food–a health risk. *Dtsch Arztebl Int*. 2012;109(4):49–55.
70. Trautvetter U, Camarinha-Silva A, Jahreis G, Lorkowski S, Glei M. High phosphorus intake and gut-related parameters—results of a randomized placebo-controlled human intervention study. *Nutr J*. 2018;17(1):23.
71. Du SF, Wang HJ, Zhang B, Zhai FY, Popkin BM. China in the period of transition from scarcity and extensive undernutrition to emerging nutrition-related non-communicable diseases, 1949–1992. *Obes Rev*. 2014;15(Suppl 1):8–15.
72. De Filippo C, Cavalieri D, Di Paola M, et al. Impact of diet in shaping gut microbiota revealed by a comparative study in children from Europe and rural Africa. *Proc Natl Acad Sci USA*. 2010;107(33):14691–14696.
73. Ou J, Carbonero F, Zoetendal EG, et al. Diet, microbiota, and microbial metabolites in colon cancer risk in rural Africans and African Americans. *Am J Clin Nutr*. 2013;98(1):111–120.
74. Wan Y, Wang F, Yuan J, et al. Effects of dietary fat on gut microbiota and faecal metabolites, and their relationship with cardiometabolic risk factors: a 6-month randomised controlled-feeding trial. *Gut*. 2019;68(8):1417–1429.
75. Zackular JP, Rogers MAM, Ruffin MT, Schloss PD. The human gut microbiome as a screening tool for colorectal cancer. *Cancer Prev Res (Phila Pa)*. 2014;7(11):1112–1121.
76. Zhao L, Zhang F, Ding X, et al. Gut bacteria selectively promoted by dietary fibers alleviate type 2 diabetes. *Science*. 2018;359(6380):1151–1156.
77. Zhang C, Yin A, Li H, et al. Dietary modulation of gut microbiota contributes to alleviation of both genetic and simple obesity in children. *EBioMedicine*. 2015;2(8):968–984.
78. Xu J, Lian F, Zhao L, et al. Structural modulation of gut microbiota during alleviation of type 2 diabetes with a Chinese herbal formula. *ISME J*. 2015;9(3):552–562.
79. Li T, Lu X, Yang X. Stachyose-enriched α-galacto-oligosaccharides regulate gut microbiota and relieve constipation in mice. *J Agric Food Chem*. 2013;61(48):11825–11831.
80. Li T, Lu X, Yang X. Evaluation of clinical safety and beneficial effects of stachyose-enriched α-galacto-oligosaccharides on gut microbiota and bowel function in humans. *Food Funct*. 2017;8(1):262–269.
81. Stoner GD, Mukhtar H. Polyphenols as cancer chemopreventive agents. *J Cell Biochem Suppl*. 1995;22:169–180.
82. Yuan X, Long Y, Ji Z, et al. Green tea liquid consumption alters the human intestinal and oral microbiome. *Mol Nutr Food Res*. 2018;62(12):e1800178.
83. Xie G, Zhou Q, Qiu C-Z, et al. Ketogenic diet poses a significant effect on imbalanced gut microbiota in infants with refractory epilepsy. *World J Gastroenterol*. 2017;23(33):6164–6171.