Diet and Gut Microbiome in Gastrointestinal Disease

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Disclosure statements: All authors declare no conflict of interest related to the current study.

Acknowledgements: The Leona B. And Harry M. Helmsley Charitable Trust, the Gastroenterology Society of Australia, Janssen, and the Australasian Gastro Intestinal Research Foundation

Abstract

The composition and function of the dynamic microbial community that constitutes the gut microbiome is continuously shaped by the host genome, mode of birth delivery, geography, life stage, antibiotic consumption, and diet. Diet is one of the most potent factors in determining microbiome integrity. Dietary factors in early life appear to substantially determine the risk of later health or disease; for example exposure to ultra-processed foods in childhood or adolescence may increase the risk of the later development of inflammatory bowel disease or colorectal cancer, thought to be mediated by modulation of the gut microbiota. Dietary factors when gut diseases are established influence symptoms and disease activity, can form a risk factor for ongoing disease, or can be used as therapy to decrease disease activity. The characterization of dietary content is currently complex and imperfect, but tools are emerging to define precisely the nature of dietary composition. Similarly the revolution in microbial analysis allows greater understanding of how diet influences microbial composition and function. Defining the interaction between diet, the gut microbiome and gastrointestinal disease is leading to radical changes in our clinical approach to these disorders.

Key words: Food, nutrition, microbiome, celiac disease, irritable bowel syndrome, functional gut disorder, FODMAP, Crohn's disease exclusion diet, exclusive enteral nutrition

Introduction

More than 100 years ago, Ellie Metchnikoff hypothesized that bacteria in the colon caused disease and suggested one could replace 'harmful microbes with useful microbes' via diet modification.[1] Recent progress in microbiology and computational biology has facilitated characterization of the microbiota, leading to a proliferation of diet and human microbiota studies. The mechanisms by which commensal organisms ferment indigestible fiber into short chain fatty acids (SCFAs), synthesize nutrients, maintain intestinal barrier function, and regulate mucosal immunity have been elucidated. It is now appreciated that commensals are not distinct from "harmful" species, but rather can become "pathogenic" when gut homeostasis is disturbed.[2] The scientific community and general population have demonstrated a keen interest in understanding how diet can promote 'gut health' and thwart disease. Foremost amongst the diseases of interest are those affecting the gastrointestinal tract.

Here we explore the impact of diet exposures and interventions on the gut microbiota in gastrointestinal disease. We summarize how diet and microbiota are defined and assessed, outline factors that influence the human gut microbiota, and discuss the relationship between diet, microbiota and gastrointestinal diseases, namely inflammatory bowel diseases, functional gut disorders, colorectal cancer, and celiac disease.

How Diet and the Microbiome are Defined and Assessed

Microbiota

The microbiome can be analyzed from fecal samples or mucosal biopsies and sequenced using 16S RNA or shotgun metagenomics. 16S RNA sequencing describes bacteria and archaea present by sequencing a specific region of DNA (the small ribosomal subunit) and relies on matching this with known sequences in publicly available databases. Shotgun metagenomics identifies microorganisms by sequencing the total DNA in a sample and can identify novel (uncharacterized) species and provide data about microbial metabolic activity.[2]

The human gut microbiota is dominated by around 160 species of bacteria that belong to six main phyla (*Firmicutes, Bacteroidetes, Actinobacteria, Proteobacteria, Fusobacteria* and

Verrumicrobia).[2] The commonest descriptive measures of the microbiota are (i) absolute and relative *abundance* of phylum, family, or genus, (ii) alpha diversity, that is the diversity of organisms, and (iii) beta diversity, a measure of microbial composition for comparison between samples. Other measures that have been used to reflect "microbial health" include *Bacteroidetes:Firmicutes* ratio, *Proteobacteria:Actinobacteria* ratio and enterotype (the predominant bacterial genera, usually *Prevotella*, *Bacteroides* or *Ruminococcus*). A higher B:F ratio, lower P:A ratio and higher overall diversity are considered more favorable markers of gut microbial health.

Diet

Diet assessment methodologies vary in their reliability, validity, suitability for individuals versus groups, and ability to capture habitual (food frequency questionnaires - FFQ, diet history) or current (24-hr recall, food record) intake. Diet can be reported as mean nutrient and food group intake, using pre-defined diet scores (e.g. dietary inflammatory index), or using unique diet patterns e.g. 'western', which is typified by high intake of processed foods, animal foods, saturated fat and low intake of fiber.[3]

General critiques of diet assessment methods have been discussed in detail elsewhere.[3] The specific complications of diet assessment in microbiota studies relate to difficulty in accurately capturing food components and eating habits that are thought to influence gut bacteria, such as fiber sub-types, pre-biotics and polyphenols, ultra-processed foods and food additives, and cooking methods.[3] Recent advances include tools to assess fiber and prebiotics, fermentable oligosaccharides, disaccharides, monosaccharides and polyols (FODMAPS), additives and ultra-processed food.

Dietary Factors that Influence the Human Gut Microbiome

Diet is just one factor that influences the composition and function of the gut microbiome, others being the host factors, birth mode, life stage, antibiotic use, and other environmental factors.[4, 5]

Host genetics impact gut microbiota heritability, metabolic potential and structure. Around 10% of taxa are thought to be heritable (e.g. *Akkermansia muciniphila,* the *Christensenellaceae* family). In some instances genetic factors have an effect on dietary tolerance or handling. Single nucleotide polymorphisms (SNPs) in the lactase gene (LCT)

are causal for lactose intolerance and LCT is associated with *Bifidobacterium* abundance.[6] SNPs in the *FUT2* (fructosyltransferase 2) and *MUC2* (mucin-2) genes negatively impact gut mucosal barrier function, nutrient bioavailability and downstream bacterial abundance.[6]

There are clear patterns of microbial succession from birth, with *Bifidobacterium* predominance on a milk based diet, followed by increasing diversity over the first year of life especially when solids are started.[4, 6] The proportion of *Bifidobacterium,* and other SCFA producing bacteria declines with age.[4]

Antibiotics alter the gut microbiota for up to four years post-exposure.[7] A decrease in diversity and richness is observed, with an expansion of pathobionts such as *Enterobacteriaceae*. Beneficial commensals (including SCFA producers such as *Faecalibacterium* and other species from *Clostridium* clusters IV and XIVa) are decreased, leading to decreases in stool butyrate.[7]

Diet patterns and individual nutrients modify the relative abundance of beneficial and pathogenic microbiota and influence overall microbiota architecture. Diets rich in plant-based foods are associated with higher microbial diversity, a predominance of anti-inflammatory, beneficial taxa and a lower abundance of pathobionts when compared to Western diets. Microbiota differences exist between vegetarians or vegans and omnivore or carnivores, likely related to fiber and saturated fat intake.[8, 9] Fermentation of fiber is an important pathway for SCFA production.[8] Conversely, saturated animal fat is associated with an increase in pro-inflammatory Lipopolysaccharide (LPS), which has been shown to expand populations of bile-tolerant pathobionts such as *Bilophila*.[8, 9]

The gut microbiota is subject to temporal shifts based on dietary pattern. Such changes can occur rapidly. David [9] et al demonstrated major, but reversible, microbiota alterations in a subject who was a lifelong vegetarian after consuming an animal-based diet for five days. In the same study ten subjects demonstrated significantly altered beta diversity within one day when introducing an animal-based diet. This microbiota-diet plasticity may be an evolutionary adaptation to seasonal food abundance.[9] This work also demonstrated that some foodborne bacterial species can survive intestinal transit, and may transiently populate the gut.[9]

Diet, Microbiota and Inflammatory Bowel Diseases

Microbial diversity is reduced, and composition changed, in Crohn's disease and ulcerative colitis[10] (Table 1). The microbiome is central to the initiation of a proinflammatory milieu in inflammatory bowel diseases (IBD), in particular Crohn's disease.[11]

Diet and IBD risk

Several epidemiological studies have reported an association between the intake of specific nutrients, foods and food patterns and the risk of developing IBD (Table 2).[12] A large population-based study with long follow-up has demonstrated that that ultra-processed food intake was associated with the development of Crohn's disease.[13]

IBD incidence is highest in Western, industrialized nations. Immigrants from countries of low IBD incidence to countries of high IBD incidence have an increased risk of developing IBD later in life. The younger the age of immigration the greater the risk, suggesting that environmental factors in childhood or adolescence have the greatest impact on the microbiota.[14]

Dietary Treatment in Crohn's disease

In the 1970's, elemental liquid nutrition combined with whole food exclusion was shown to produce clinical improvement that could not be explained by correction of malnutrition alone. In 1984, O'Moráin, et al. [15] published the first controlled trial demonstrating that an elemental diet was equal to corticosteroids for inducing remission of Crohn's disease. Multiple randomized controlled trials have since confirmed that exclusive enteral nutrition (EEN) is of therapeutic benefit in pediatric and adult patients.[16] Initial hypotheses explaining the efficacy of EEN related to removal of 'antigenic' food proteins and alteration of gut flora but more recently the effects of EEN on the gut microbiota have been described.[17] Paradoxically, EEN reduces anti-inflammatory *Faecalibacterium* and increases abundance of pro-inflammatory *Alistipes* (Table 3). It has been suggested that these shifts allow for microbial restoration in the long-term.[17]

Tolerance and compliance are major practical limitations of EEN use, prompting the development of food-based dietary therapies in IBD (Table 4). A diet composed of food constituents mimicking the composition of EEN (CD-Treat) has demonstrated microbial

composition and diversity shifts in the same direction as EEN in healthy volunteers, and clinical remission in 3 of 5 paediatric patients.[18]

Historically, partial enteral nutrition (PEN) has failed to reduce inflammation, leading researchers to surmise that the removal of specific foods drives the success of EEN. Subsequently, Levine, et al. [19] developed the Crohn's Disease Exclusion Diet (CDED), a diet which restricts whole food intake that has been used in combination with a liquid polymeric diet. A key principle of the diet is the exclusion of food additives and restriction of red meat, dairy and wheat, dietary components that are postulated to have a negative impact on the microbiome, host intestinal barrier function and immunity.[19] Other principles of the diet are mandatory daily intake of potatoes, apples, and bananas, which provide fiber and resistant starch that are fermented by *Bifidobacterium* and other saccharolytic species to produce butyrate.[8] The CDED with PEN was equally effective as EEN for inducing remission in mild-moderate luminal Crohn's Disease.[19] The CDED with PEN and EEN both led to a decrease in *Proteobacteria* at week 6, but these changes were only maintained at week 12 on CDED with PEN. Non-responders at week 6 had less microbial changes than responders. It is unclear which components of the diet are responsible for these changes.

The major effects of food additives on microbial composition and function are becoming increasingly clear. Chassaing, et al. [20] demonstrated that the emulsifiers carboxymethylcellulose and polysorbate-80 contribute to intestinal hyperpermeability and inflammation via effects on the microbiota, including expansion of mucolytic *Ruminococcus gnavus*, and the suppression of *Faecalibacterium prausnitzii.* Further deleterious effects of these and other additives have since been described.[21] Crohn's disease patients appear to have had, and continue to have, a higher intake of food additives[22] and ultra-processed foods[23] compared to a range of control subjects across multiple countries.

The Mediterranean and specific carbohydrate diets (SCD) have also been evaluated in cohorts with IBD. Both diets are low in food additives. A recent RCT comparing the two approaches in mild-moderate Crohn's disease reported clinical efficacy without demonstrable benefit in objective measures of inflammation; richness and Shannon's diversity index were comparable across diet arm and did not change across the course of the study.[24] One postulated mechanism for the benefit of the SCD is a microbial change due to the removal of disaccharides and polysaccharides, purported to cause bacterial overgrowth, yet there is limited evidence of microbial changes on the diet. Clinical benefits from the SCD may relate to symptom improvement associated with reduced FODMAP

intake.[25] The impact Mediterranean food components on the microbiota is summarised in Table 4.

Dietary Treatment in Ulcerative colitis

Dietary approaches in ulcerative colitis have received less attention than in Crohn's disease. In the 1960s Truelove [26] proposed that ulcerative colitis was provoked by milk. More recently milk-fat, but not safflower oil, has been shown to promote the expansion of *Bilophila wadsworthia*, a sulphate-reducing bacteria that induces colitis in susceptible mice.[27] Milk fat promotes taurine-conjugation of hepatic bile acids, which increases the availability of organic sulfur and subsequent production of H_2S, a mediator of inflammation with the potential to disrupt mucosal integrity.[27] Animal proteins, sulphite preservatives, and the gelling agent carrageenan are other dietary sources of sulfur.[28] A small RCT found carrageenan capsules led to higher clinical relapse rates than placebo.[29] Plant-based diets have also been shown to prevent relapse in ulcerative colitis.[30]

Diet, Gut Microbiota and Functional Gastrointestinal Disorders

The pathophysiologic mechanisms which underlie symptoms in functional gut disorders include altered motility, sensory disturbances such as visceral hypersensitivity dysregulated gut immune function and the gut microbiome.[31]

Studies which have attempted to categorize the microbiota in functional gut disorders have reported varied results, likely owing to heterogenous symptom profiles[32] and challenges in controlling for contributory factors such as patient's habitual diet.[33] A greater abundance of *Proteobacteria* is the most consistent change reported at the phylum level in Irritable Bowel Syndrome (IBS)[32] (Table 1). Clinical symptoms may also influence the gut microbiota. A longitudinal multi-omics study revealed that patients who describe symptoms in a "flare" demonstrated altered microbial composition and those with "severe" IBS were more likely to have increased abundance of *Lactobacillus* spp.[33]

Patients are aware that diet contributes to their IBS symptoms. Traditional dietary advice and first-line diet strategies focus on meal size, fiber modification, and avoidance of deep-fried foods, 'gas-producing foods' (e.g. cabbage) and gut irritants (e.g. alcohol, caffeine,

spicy foods).[31] Limiting poorly absorbed, fermentable carbohydrates (deemed - FODMAPs) has been shown in controlled trials to be effective in reducing IBS symptoms in 50-80% of individuals.[34]

Colonic, bacterial fermentation of certain FODMAPs leads to similar levels of gas production and luminal distention in individuals with and without IBS. It is theorized that those with IBS develop symptoms due to visceral hypersensitivity.[31] In a placebo controlled trial, fecal transplant using a single well characterized “super-donor” significantly reduced IBS symptom scores[35], supporting a role of the microbiota in symptom induction. A recent study found no associations between habitual diet of IBS sufferers and gut microbiota when correcting for multiple comparisons, further raising the possibility that personalized and disease-specific responses alter microbiota-diet interactions in these patients.[36]

The altered microbiota in IBS has been postulated to result in abnormal levels of intestinal fermentation as demonstrated by altered lactase-metabolizing and hydrogen-consuming bacteria.[24] Fecal volatile compounds, a metabolomic measure, have also been shown to be predictor of response to a FODMAP diet.[37]

Although producing symptomatic benefits, low FODMAP diets have been shown to lower levels of Bifidobacteria, and in some studies *Faecalibacterium prausnitzii*, *Clostridium* cluster IV*, Faecalibacterium prausnitzii, Firmicutes and Clostridiales*[38] (Table 5). The clinical relevance of these microbial changes is not known but concerns over a reduction in supposed beneficial species has led to attempts to ameliorate these effects. Staudacher, et al. [39] demonstrated that probiotic supplementation restored *Bifidobacterium* in individuals with IBS. Further, probiotics have been shown to increase the abundance of *Lactobacillus*, *Streptococcus* and the *Lactobacillaceae* family, with no effect on *Bifidobacterium*.[36] Some microbiota responses to low FODMAP diets may be explained by altered intake of food components that co-occur in high FODMAP foods and impact the gut microbiome (e.g. polyphenols) but are rarely measured in diet studies.

Despite associations between gut microbiota and food additives[21], no studies of these factors have been conducted in patients with functional gut disorders, thus their contribution to pathophysiology and symptoms is not known.

Diet, Microbiota and Colorectal Cancer

Colorectal cancer is the 3rd commonest cancer and 2nd most common cause of cancer death globally.[40]

A wide range of epidemiological studies have demonstrated an association between a Western diet, in particular high meat, animal fat, alcohol and low fiber intake, and the development of colorectal cancer (Table 2).[40] Epidemiological studies often enroll participants in middle age. A recent prospective investigation of sugar-sweetened beverage intake during adolescence found that frequent (>2 serves/day) consumption was associated with a 32% greater risk of developing early onset colorectal cancer.[41] The authors proposed that dysbiosis induced by high fructose corn syrup, a common sweetener in soft drinks in North America, may promote colorectal cacogenesis.[41]

While individuals who migrate from Asia, Europe and the Middle East to North America have lower colorectal cancer incidence than non-immigrants, increased length of stay increases cancer risk.[42]

Alterations in the microbiota of individuals with colorectal cancer include decreased overall diversity, decreased anti-inflammatory microbes including *Clostridia* spp.[40], and increases in the pro-inflammatory genera *Fusobacterium, Porphyromonas, Peptostreptococcus* and *Prevotella,* and mucosal associated *Escherichia coli*[40, 43] (Table 1). Enterotoxigenic *Bacteroides fragilis* promotes intestinal inflammation and is enriched in tumor tissue, intestinal mucosa and fecal samples of patients with colorectal cancer, particularly when advanced tumors are present. In murine models, *Escherichia coli* invades the mucosa and becomes an intracellular microbe with oncogenic potential.[40]

The association between the intestinal microbiome and development and progression of colorectal cancer may be mediated by nutritional components that modulate chronic gut inflammation (Table 2). *Fusobacterium* is the best defined microbe with a link to colorectal cancer and diet.[31] Diets high in whole grains and dietary fibers significantly decrease the amount of *Fusobacterium nucleatum* in subjects with colorectal cancer.[40444444] In a similar fashion, susceptibility to *Escherichia coli* and intestinal inflammation is enhanced by consumption of a Western diet[43] and polysorbate-80 in animal models.[21]

Ketogenic and fasting diets have been proposed as adjunct therapies to chemotherapy and radiotherapy. Animal models demonstrate that starvation can reduce cancer incidence and

severity. Ketone bodies reduce proinflammatory Th17 cells; however, ketogenic diets have mostly “unfavorable” effects on the gut microbiome (Table 4).[45]

Diet, Gut Microbiota and Celiac Disease

Celiac disease is characterized by T-cells mounting a pro-inflammatory response to gluten (specifically, gliadin in wheat and prolamines in barley and rye), resulting in small bowel inflammation and villous atrophy. In celiac disease, gluten also stimulates zonulin release, a disrupter of intestinal membrane tight junctions. The treatment for patients with celiac disease is strict adherence to a gluten free diet.[46]

Longitudinal studies have revealed that individuals with celiac disease have unique microbial signatures that precede disease diagnosis (presence of inflammatory *Dialister invisus*, *Parabacteroides* spp., *Lachnospiraceae*, and decreased abundance of anti-inflammatory *Streptococcus thermophilus*, *Faecalibacterium prausnitzii*, and *Clostridium clostridioforme).* Altered composition is also apparent in active disease when a gluten free diet is not adhered to (decreased diversity of *Lactobacillus* and *Bifidobacterum* spp.*).*[47] Further microbial alterations occur in response to a gluten free diet (increases in abundance of *Firmicutes*, *Streptococcus spp.*, *Prevotella*, *Actinobacteria* and *Gammaproteobacteria).* *C*omplete ‘normalization’ of gut microbiota does not occur even when inflammation is not present.[46]

Persistence of microbial disturbances in individuals on a gluten free diet may be partially related to restriction of fructan-rich and gluten-rich grains, which promote the growth of beneficial, butyrate producing species.[8] The gut microbiota may also have a role in presence of persistent gastrointestinal symptoms in patients with celiac disease despite a gluten free diet. Individuals with persistent gastrointestinal symptoms have increased abundance of *Proteobacteria* and decreased abundance *of Firmicutes* and *Bacteroidetes*.[46]

Fermented (Probiotic) Foods

Fermentation of foods and beverages, via controlled microbial growth, is a an ancient, traditional method of food preservation that leads to the production of ‘dietary probiotics’ – foodstuffs that contain live bacterial cultures. The impact of fermented foods, such as yoghurt[48], kefir, kimchi, miso, sauerkraut, sourdough bread, on gut microbiota and

gastrointestinal health have been extensively reviewed elsewhere[49]. Evidence for the impacts of most fermented foods are limited by small study numbers and lack of ability to tease out impact of live bacterial cultures from other food ingredients. Notable findings that related to gastrointestinal include: (1) kefir led to increased stool *Lactobacillus* in patients with Crohn's disease (2) both sauerkraut and pasteurized cabbage (no live microbes) led to a reduction in IBS symptoms, but not to microbiota changes, (3) sauerkraut may increase risk of laryngeal cancers, although conflicting results have been reported (4)

Human studies on tempeh, kombucha

The Mediterranean diet includes frequent consumption of yoghurt, a dietary probiotic. Yoghurt has also been associated with a decreased risk of developing colorectal cancer (Table 2).

Prebiotic Foods

Prebiotic foods contain dietary fibers that are fermented by commensal gut species to produce beneficial bacteria[50]. Prebiotic rich foods include apples, bananas, barley, garlic, leek, oats, and onions. Grains, vegetables, onion and garlic are mainstays of the Mediterranean diet that have beneficial effects on the gut (Table 4). Apples and bananas are mandatory foods in the CDED and promote the growth of butyrate-producing species (Table 4).

Recommendations and Directions for Future Research

Patients with gastrointestinal d**i**sorders are prone to dietary restriction and malnutrition, and should be encouraged to consume diets that are as liberal and diverse as possible, with special attention given to plant-based foods that promote abundance of beneficial bacteria.

Inflammatory bowel disease

Diet strategies to modify the microbiota towards a profile that may reduce risk of IBD should be considered, especially in individuals with family history of IBD. Therapeutic diets (EEN, CDED) can be used to treat inflammation via microbiota manipulation, and to treat symptoms in existing disease. Future research should focus on further elucidating the mechanisms by which these diets exert their effects, and on developing evidence-based diet strategies for ulcerative colitis.

Functional gut disorders

Low FODMAP diets appear to be more successfully implemented when supervised by dietitians familiar with this strategy. Adequate fiber, polyphenol and resistant starch intake should be addressed.

Future research may address the clinical implications of microbial differences in IBS and the longer term impact of dietary manipulation.

Colorectal cancer

Dietary strategies to modify the microbiota towards a profile that may reduce risk of colorectal cancer should be considered, especially in individuals with family history of colorectal cancer.

Future research should assess the efficacy of therapeutic diets, for example their efficacy in modifying microbial factors such as *Fusobacterium* abundance.

Celiac disease

Dietary strategies to optimize microbiota for those on a life-long gluten-free diet should be encouraged. These include ensuring adequate consumption of fiber, plant foods, fermented foods and non-gluten containing grains. Future research should focus on possible negative effects of ultra-processed gluten-free foods on the gut microbiota.

Conclusions

Diet shapes the gut microbiome in a manner that influences the development and activity of gastrointestinal disease. Animal and longitudinal human studies support the role of an altered microbiome as a cause of disease. Interindividual difference in the microbiota remains greater than intraindividual variation, highlighting a complex interplay of genes and environment in microbiota development, structure and function, and supporting the idea that personalized, disease-specific diet-microbiota responses occur.

Diet-induced microbiota responses partly explain how diet patterns modulate disease risk and the mechanisms through which diet modifies inflammation.

While the gut microbiota can be described using several broad categories a clear definition of a ‘healthy’ microbiota does not exist. In the future, diet will be harnessed to prevent gastrointestinal diseases with early life and childhood being critical periods of gut microbiota development. Microbial treatments will carry lower risk than pharmacological intervention.

References

1. Podolsky SH. Metchnikoff and the microbiome. The Lancet. 2012;380(9856):1810-1.
2. Young VB. The role of the microbiome in human health and disease: an introduction for clinicians. BMJ. 2017;356:j831.
3. Johnson AJ, Zheng JJ, Kang JW, Saboe A, Knights D, Zivkovic AM. A guide to diet-microbiome study design. Frontiers in Nutrition. 2020;7:79.
4. Yatsunenko T, Rey FE, Manary MJ, Trehan I, Dominguez-Bello MG, Contreras M, et al. Human gut microbiome viewed across age and geography. Nature. 2012;486(7402):222-7.
5. Bokulich NA, Chung J, Battaglia T, Henderson N, Jay M, Li H, et al. Antibiotics, birth mode, and diet shape microbiome maturation during early life. Sci Transl Med. 2016;8(343):343ra82.
6. Hall AB, Tolonen AC, Xavier RJ. Human genetic variation and the gut microbiome in disease. Nature Reviews Genetics. 2017;18(11):690-9.
7. Zimmermann P, Curtis N. The effect of antibiotics on the composition of the intestinal microbiota-a systematic review. Journal of Infection. 2019;79(6):471-89.
8. Zmora N, Suez J, Elinav E. You are what you eat: diet, health and the gut microbiota. Nature reviews Gastroenterology & hepatology. 2019;16(1):35-56.
9. David LA, Maurice CF, Carmody RN, Gootenberg DB, Button JE, Wolfe BE, et al. Diet rapidly and reproducibly alters the human gut microbiome. Nature. 2014;505(7484):559-63.
10. Gevers D, Kugathasan S, Denson LA, Vázquez-Baeza Y, Van Treuren W, Ren B, et al. The treatment-naive microbiome in new-onset Crohn's disease. Cell host & microbe. 2014;15(3):382-92.
11. Nagao-Kitamoto H, Shreiner AB, Gillilland III MG, Kitamoto S, Ishii C, Hirayama A, et al. Functional characterization of inflammatory bowel disease–associated gut dysbiosis in gnotobiotic mice. Cellular and molecular gastroenterology and hepatology. 2016;2(4):468-81.
12. Piovani D, Danese S, Peyrin-Biroulet L, Nikolopoulos GK, Lytras T, Bonovas S. Environmental Risk Factors for Inflammatory Bowel Diseases: An Umbrella Review of Meta-analyses. Gastroenterology. 2019;157(3):647-59.e4.
13. Lo CH, Khandpur N, Rossato SL, Lochhead P, Lopes EW, Burke KE, et al. Ultra-processed Foods and Risk of Crohn's Disease and Ulcerative Colitis: A Prospective Cohort Study. Clin Gastroenterol Hepatol. 2021.
14. Benchimol EI, Mack DR, Guttmann A, Nguyen GC, To T, Mojaverian N, et al. Inflammatory Bowel Disease in Immigrants to Canada And Their Children: A Population-Based Cohort Study. Official journal of the American College of Gastroenterology | ACG. 2015;110(4).
15. O'Moráin C, Segal AW, Levi AJ. Elemental diet as primary treatment of acute Crohn's disease: a controlled trial. Br Med J (Clin Res Ed). 1984;288(6434):1859-62.
16. Narula N, Dhillon A, Zhang D, Sherlock ME, Tondeur M, Zachos M. Enteral nutritional therapy for induction of remission in Crohn's disease. Cochrane Database of Systematic Reviews. 2018(4).
17. MacLellan A, Connors J, Grant S, Cahill L, Langille MG, Van Limbergen J. The impact of exclusive enteral nutrition (EEN) on the gut microbiome in Crohn's disease: a review. Nutrients. 2017;9(5):447.

18. Svolos V, Hansen R, Nichols B, Quince C, Ijaz UZ, Papadopoulou RT, et al. Treatment of active Crohn's disease with an ordinary food-based diet that replicates exclusive enteral nutrition. Gastroenterology. 2019;156(5):1354-67. e6.
19. Levine A, Wine E, Assa A, Boneh RS, Shaoul R, Kori M, et al. Crohn's disease exclusion diet plus partial enteral nutrition induces sustained remission in a randomized controlled trial. Gastroenterology. 2019;157(2):440-50. e8.
20. Chassaing B, Koren O, Goodrich JK, Poole AC, Srinivasan S, Ley RE, et al. Dietary emulsifiers impact the mouse gut microbiota promoting colitis and metabolic syndrome. Nature. 2015;519(7541):92-6.
21. Levine A, Boneh RS, Wine E. Evolving role of diet in the pathogenesis and treatment of inflammatory bowel diseases. Gut. 2018;67(9):1726-38.
22. Trakman GL, Lin WY, Hamilton AL, Wilson-O'Brien A, Stanley A, Or L, et al. Sa513 FOOD AS A RISK FACTOR FOR THE DEVELOPMENT AND PERPETUATION OF CROHN'S DISEASE. AN INTERNATIONAL CASE-CONTROL STUDY OF FOOD AND FOOD ADDITIVE INTAKE FROM BIRTH TILL NOW. THE ENIGMA STUDY. Gastroenterology. 2021;160(6):S-530.
23. Narula N, Wong EC, Dehghan M, Mente A, Rangarajan S, Lanas F, et al. Association of ultra-processed food intake with risk of inflammatory bowel disease: prospective cohort study. bmj. 2021;374.
24. Lewis JD, Sandler RS, Brotherton C, Brensinger C, Li H, Kappelman MD, et al. A Randomized Trial Comparing the Specific Carbohydrate Diet to a Mediterranean Diet in Adults With Crohn's Disease. Gastroenterology. 2021;161(3):837-52.e9.
25. Cox SR, Lindsay JO, Fromentin S, Stagg AJ, McCarthy NE, Galleron N, et al. Effects of low FODMAP diet on symptoms, fecal microbiome, and markers of inflammation in patients with quiescent inflammatory bowel disease in a randomized trial. Gastroenterology. 2020;158(1):176-88. e7.
26. Truelove SC. Ulcerative colitis provoked by milk. Br Med J. 1961;1(5220):154-60.
27. Devkota S, Wang Y, Musch MW, Leone V, Fehlner-Peach H, Nadimpalli A, et al. Dietary-fat-induced taurocholic acid promotes pathobiont expansion and colitis in Il10-/- mice. Nature. 2012;487(7405):104-8.
28. Costello SP, Hughes PA, Waters O, Bryant RV, Vincent AD, Blatchford P, et al. Effect of Fecal Microbiota Transplantation on 8-Week Remission in Patients With Ulcerative Colitis: A Randomized Clinical Trial. JAMA. 2019;321(2):156-64.
29. Bhattacharyya S, Shumard T, Xie H, Dodda A, Varady KA, Feferman L, et al. A randomized trial of the effects of the no-carrageenan diet on ulcerative colitis disease activity. Nutr Healthy Aging. 2017;4(2):181-92.
30. Chiba M, Nakane K, Tsuji T, Tsuda S, Ishii H, Ohno H, et al. Relapse prevention in ulcerative colitis by plant-based diet through educational hospitalization: A single-group trial. The Permanente Journal. 2018;22.
31. Simrén M. Diet as a therapy for irritable bowel syndrome: progress at last. Gastroenterology. 2014;146(1):10-2.
32. Pittayanon R, Lau JT, Yuan Y, Leontiadis GI, Tse F, Surette M, et al. Gut microbiota in patients with irritable bowel syndrome—a systematic review. Gastroenterology. 2019;157(1):97-108.
33. Mars RA, Yang Y, Ward T, Houtti M, Priya S, Lekatz HR, et al. Longitudinal multi-omics reveals subset-specific mechanisms underlying irritable bowel syndrome. Cell. 2020;182(6):1460-73. e17.

34. Schumann D, Klose P, Lauche R, Dobos G, Langhorst J, Cramer H. Low fermentable, oligo-, di-, mono-saccharides and polyol diet in the treatment of irritable bowel syndrome: A systematic review and meta-analysis. Nutrition. 2018;45:24-31.
35. El-Salhy M, Hatlebakk JG, Gilja OH, Bråthen Kristoffersen A, Hausken T. Efficacy of faecal microbiota transplantation for patients with irritable bowel syndrome in a randomised, double-blind, placebo-controlled study. Gut. 2020;69(5):859-67.
36. Staudacher HM, Scholz M, Lomer MC, Ralph FS, Irving PM, Lindsay JO, et al. Gut microbiota associations with diet in irritable bowel syndrome and the effect of low FODMAP diet and probiotics. Clinical Nutrition. 2021;40(4):1861-70.
37. Rossi M, Aggio R, Staudacher HM, Lomer MC, Lindsay JO, Irving P, et al. Volatile Organic Compounds in Feces Associate With Response to Dietary Intervention in Patients With Irritable Bowel Syndrome. Clinical Gastroenterology and Hepatology. 2018;16(3):385-91.e1.
38. Reddel S, Putignani L, Del Chierico F. The Impact of Low-FODMAPs, Gluten-Free, and Ketogenic Diets on Gut Microbiota Modulation in Pathological Conditions. Nutrients. 2019;11(2):373.
39. Staudacher HM, Lomer MC, Farquharson FM, Louis P, Fava F, Franciosi E, et al. A diet low in FODMAPs reduces symptoms in patients with irritable bowel syndrome and a probiotic restores bifidobacterium species: a randomized controlled trial. Gastroenterology. 2017;153(4):936-47.
40. Song M, Chan AT, Sun J. Influence of the gut microbiome, diet, and environment on risk of colorectal cancer. Gastroenterology. 2020;158(2):322-40.
41. Hur J, Otegbeye E, Joh H-K, Nimptsch K, Ng K, Ogino S, et al. Sugar-sweetened beverage intake in adulthood and adolescence and risk of early-onset colorectal cancer among women. Gut. 2021.
42. Flood DM, Weiss NS, Cook LS, Emerson JC, Schwartz SM, Potter JD. Colorectal cancer incidence in Asian migrants to the United States and their descendants. Cancer Causes & Control. 2000;11(5):403-11.
43. Agus A, Denizot J, Thévenot J, Martinez-Medina M, Massier S, Sauvanet P, et al. Western diet induces a shift in microbiota composition enhancing susceptibility to Adherent-Invasive E. coli infection and intestinal inflammation. Scientific Reports. 2016;6(1):19032.
44. Mehta RS, Nishihara R, Cao Y, Song M, Mima K, Qian ZR, et al. Association of Dietary Patterns With Risk of Colorectal Cancer Subtypes Classified by Fusobacterium nucleatum in Tumor Tissue. JAMA Oncol. 2017;3(7):921-7.
45. Shirdarreh M, Sadeghi Y, Rahimi T. The Impact of Ketogenic Diet on Colorectal Cancer Progression and the Co-evolution of Gut Microbiota: A Research Protocol. Undergraduate Research in Natural and Clinical Science and Technology Journal. 2021:1-6.
46. Caio G, Lungaro L, Segata N, Guarino M, Zoli G, Volta U, et al. Effect of gluten-free diet on gut microbiota composition in patients with celiac disease and non-celiac gluten/wheat sensitivity. Nutrients. 2020;12(6):1832.
47. Leonard MM, Valitutti F, Karathia H, Pujolassos M, Kenyon V, Fanelli B, et al. Microbiome signatures of progression toward celiac disease onset in at-risk children in a longitudinal prospective cohort study. Proceedings of the National Academy of Sciences. 2021;118(29).
48. Aryana KJ, Olson DW. A 100-Year Review: Yogurt and other cultured dairy products. Journal of Dairy Science. 2017;100(12):9987-10013.

604 49. Dimidi E, Cox SR, Rossi M, Whelan K. Fermented Foods: Definitions and
605 Characteristics, Impact on the Gut Microbiota and Effects on Gastrointestinal Health and
606 Disease. Nutrients. 2019;11(8):1806.
607 50. Albenberg LG, Wu GD. Diet and the intestinal microbiome: associations, functions,
608 and implications for health and disease. Gastroenterology. 2014;146(6):1564-72.
609 51. Graf D, Di Cagno R, Fåk F, Flint HJ, Nyman M, Saarela M, et al. Contribution of diet to
610 the composition of the human gut microbiota. Microbial Ecology in Health and Disease.
611 2015;26(1):26164.
612 52. Bond T, Derbyshire E. Tea compounds and the gut microbiome: findings from trials
613 and mechanistic studies. Nutrients. 2019;11(10):2364.
614 53. Singh RK, Chang H-W, Yan D, Lee KM, Ucmak D, Wong K, et al. Influence of diet on
615 the gut microbiome and implications for human health. Journal of Translational Medicine.
616 2017;15(1):73.
617 54. Gomes J, Costa J, Alfenas R. Could the beneficial effects of dietary calcium on obesity
618 and diabetes control be mediated by changes in intestinal microbiota and integrity? British
619 Journal of Nutrition. 2015;114(11):1756-65.
620 55. Suzuki Y, Ikeda K, Sakuma K, Kawai S, Sawaki K, Asahara T, et al. Association between
621 yogurt consumption and intestinal microbiota in healthy young adults differs by host
622 gender. Frontiers in microbiology. 2017;8:847.
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Table 1: Microbial Patterns in Gastrointestinal Disease

Disease	Putative Microbial Patterns in Select Studies	
Celiac disease	↑ *Dialister invisus*, *Parabacteroides* spp., *Lachnospiraceae*, ↓ *Streptococcus thermophilus, Faecalibacterium prausnitzii,* and *Clostridium clostridioforme* (pre-disease) ↓ *Lactobacillus and Bifidobacterium* (active disease)	46, 47
Colorectal cancer	↓ *Clostridia* spp. ↑ *Fusobacterium nucleatum, Bacteroides fragilis and Escherichia coli*	40
Irritable Bowel Syndrome	↑ *Proteobacteria* (4 studies; 2 studies no change) ↑ *Bacteroidetes* (2 studies; 4 studies no change) ↑ *Actinobacteria* (1 studies; 3 or more studies no change) ↑ *Firmicutes* (1 studies; 3 or more studies no change) ↑ *Lactobacillus* spp. (severe)	32, 33
Crohn's Disease	↓ Overall Diversity ↓ *Firmicutes, Ruminococcus, Lachnospiraceae and Roseburia* ↑ *Pasturellaceae, Veillonellaceae, Neisseriaceae, Fusobacteriaceae, and Enterobacteriaceae*	10
Ulcerative Colitis	↓ Overall Diversity ↓ *Firmicutes* ↑ *Proteobacteria,* ↑ *Alistipes massiliensis, Ruminococcus gnavus, Escherichia coli, Helicobacter and Campylobacter* spp.	15

Crohn' disease changes are based on the largest study of treatment-naive, newly diagnosed Crohn's disease in pediatric patients. IBS changes are variable and are surmised from a systematic literature review and predominately reported here at the phylum level.

Table 2: Disease Risk Related to Diet May be Mediated by the Changes in the Gut Microbiome

Inflammatory bowel disease*	Fiber (CD) ↑ butyrate which ↑ T regulatory cells and ↓ colonic stem cell proliferation; ↑ *Bifidobacterium spp. and Lactobacillus spp.*	[51]
	Tea (UC) ↑ microbial diversity, *Bifidobacterium,* and *Bacteroidetes*: *Firmicutes* ratio	[52]
	Fruit (CD, UC) and Vegetables[+] (UC) contain fiber and polyphenols, which ↑ microbial diversity, beneficial *Bifidobacterium* and *Lactobacillus*	[51]
	Soft drink and sucrose (UC) [a] ↓ beneficial *Bacteroidetes*	[53]
	Ultra-processed foods (CD) ↑ additives, saturated fat, sugar, and low in fiber	[13, 23]
Colorectal cancer*	Fiber ↑ butyrate which ↑ T regulatory cells and ↓ colonic stem cell proliferation	[51]
	Calcium ↑gastric acid secretion and colonic pH (via bile acid precipitation) and therefore ↓secondary bile acids and non-esterified fatty acids. ↑ GLP2 and therefore ↑ tight junction gene expression and intestinal permeability and ↓ lipopolysaccharide and bacterial translocation	[54]
	Yoghurt ↑ beneficial *Bifidobacterium* spp.	[55]
	Wholegrains ↑ beneficial *Bifidobacterium* spp. ↓ *Fusobacterium nucleatum*	[51]
	Red meat and animal fat ↑ 7α dehydroxylating bacteria, which produce cytotoxic secondary bile acids; ↑ *Bacteroides and Bilophilia;* [a]source of haem iron which induces cytotoxic stress	[51]
	Alcohol (in excess) ↑ pro-inflammatory *Proteobacteria* and ↓ *Bacteroidetes (*including *Verrucomicrobia)* and *Firmicutes (*including *Enterobacteriaceae).* ↑ intestinal permeability, lipopolysaccharide and bacterial translocation	
	Sugar sweetened beverages, prepared with high-fructose-corn syrup in North America, which ↓ Firmicutes ↑ Bacteroidetes, ↑ Ruminococcus; [a]epithelial barrier dysfunction and increased intestinal permeability	

Notes: Protective factors are green; Risk factors are orange

Fiber, fruit, vegetables are typically low in a Western diet; Additives, saturated fat and animal protein are typically high in a Western diet.

* Risk and protective factors for colorectal cancer and IBD predominately derived from umbrella review of systematic reviews and meta-analyses; other nutrients, foods and diet patterns have also been implicated in individual studies.

Table 3: Gut Microbiome Exhibits "Paradoxical" Responses to Therapeutic Diets		
Exclusive Enteral Nutrition (RCT evidence-Crohn's Disease)	↓ Overall diversity ↓ *Bacteroidetes* ↓ *Bacteroides* ↓ *Faecalibacterium, Bifidobacterium, Ruminococcus* ↓ *and* ↑ *Prevotella* ↑ *Lactococcus* ↑ *Alistipes* Remission positively associated with *Akkermansia muciniphila*, *Bacteroides*, *Lachnospiraceae,* and *Ruminococcaceae*	[17]
Ketogenic diet (Suggested - Colorectal Cancer)	Ketone bodies ↓ Th17 proinflammatory T cells. Negative effects also seen. ↓ Diversity ↑ *Bacteroidetes* and ↓ *Proteobacteria*; ↓ *Firmicutes* and *Actinobacteria* ↑ *Bacteroides*, *Bifidobacterium* and *Prevotella*; ↓*Cronobacter* ↑ *Desulfovibrio*	[38, 45]

Table 4: Gut Microbiome Is a Target for Therapeutic Interventions		
Crohn's Disease Exclusion Diet (RCT evidence-Crohn's Disease)	↑ Soluble fiber and resistant starch [+] ↑ anti-inflammatory butyrate production	51
	↓ Additives[a], ↑ intestinal permeability, bacterial translocation (e.g. polysorbate-80), bacterial adherence to mucosa (e.g. carboxymethylcellulose) and negatively alter microbial composition (e.g. Carrageenan - ↓ *Bacteroidetes* and *Verrumicorbia,* and aspartame ↑ *Enterobacteriaceae)*	19, 21
	↓ Milk fat[a] and Saturated fat[a] ↓ microbial diversity and ↑ H_2S production, which, in excess, can break disulphide bonds of the intestinal epithelium and ↓ gut permeability and ↑ gut inflammation	
	↓ Red meat source of saturated fat and sulfur	
Sulphide-reducing diet (Suggested - Ulcerative Colitis)	↑ Soluble fiber and resistant starch[†] ↓ H_2S production as metabolised preferentially over animal proteins and other sources of sulfur	51
	↓ Sulfhur[a] (from animal protein, sulphated food additives e.g. carrageenan and sulphite preservatives) are fermented by sulphate-reducing bacteria (e.g. *Bilophila wadsworthia)* and ↑ H_2S, which, in excess, can break disulphide bonds of the intestinal epithelium and ↓ gut permeability and ↑ gut inflammation	27, 28
	↓ Saturated fat ↑ taurine-conjugation of bile acids, which ↑sulfur	
Mediterranean diet (Suggested – IBD)	Soluble fiber and resistant starch[+] ↑ butyrate	51
	Fruit and Vegetables contain fiber and polyphenols, which ↑ microbial diversity, beneficial *Lactobacillus* and *Bifidobacterium*	
	Yoghurt ↑ beneficial *Bifidobacterium*	
	Wholegrains ↑ beneficial *Bifidobacterium*	
	Nuts ↑ Overall diversity and capacity for butyrate production	
	Red wine (consumed in moderation) ↑ *Bifidobacterium, Enterococcus, Eggerthella lenta*	
	Extra virgin olive oil, rich in polyphenols, ↑ Overall diversity, ↑ butyrate producing *Clostridium XIVa; ↑ Bifidobacterium* spp. *and ↑ Parascardovia.*	
	Red meat and animal fat ↑ 7α dehydroxylating bacteria, which produce cytotoxic secondary bile acids	
Notes: Protective factors are green; Suggested adverse factors are orange		

Table 5: Gut Microbiome May be Impacted by Therapeutic Diets Clinical Effects Not Known	
↓ *Firmicutes, Streptococcus, Prevotella, Actinobacteria* and *Gammaproteobacteria* ↓ *Lactobacillus*, *Enterococcus* and *Bifidobacterium*; ↑ *Bacteroides*, *Staphylococcus*, *Salmonella*, *Shigella* and *Klebsiella*	38, 46
↓ *Bifidobacterium* +/- ↓ *Clostridium cluster IV* ↓ *Faecalibacterium prausnitzii,* +/- ↓ *Firmicutes* ↓ *Clostridiales* ↑ *Akkermansia muciniphila*	*38*
* Mechanism of action malabsorption of fermentable carbohydrates; diet not designed to alter microbiota **Mechanism of action autoimmune activation; diet not designed to alter microbiota. [a]based on animal studies	

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