

# The ketogenic diet: its impact on human gut microbiota and potential consequent health outcomes: a systematic literature review

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## ABSTRACT

**Aim:** This systematic review examined the diet's impact on the human gut microbiota to identify potential consequent health outcomes.

**Background:** The extreme macronutrient profile of the ketogenic diet (KD) instigates compositional shifts in the gut's microbial community.

**Methods:** In this systematic literature review, an evidence-based and methodical approach was undertaken, which involved systematic searches of the Medical Literature Analysis and Retrieval System Online (MEDLINE), PubMed and Cumulative Index to Nursing and Allied Health Literature (CINAHL) databases, generating a total of 263 relevant research papers. Following the application of inclusion and exclusion criteria, eight papers were deemed suitable for inclusion. These papers were critically appraised using a checklist tool adapted from the National Institute of Care and Excellence (NICE). The findings were analysed using a simplified thematic analysis.

**Results:** The results provide strong evidence for a persistent reduction in *Bifidobacterium* abundance following KD adherence. A reduced abundance of key *Firmicutes* butyrate-producing bacteria was found to be a likely impact, although two studies with extended intervention periods indicate this may be time-limited. Studies investigating short-chain fatty acids (SCFA's) indicate KD reduces total faecal SCFA's, acetate, and butyrate.

**Conclusion:** Changes to microbial communities resulting from KD adherence are potentially detrimental to colonic health. The persistent reduction in *Bifidobacterium* abundance was concerning, with obesity, type-2 diabetes, and depression highlighted as potential consequent risks. For nutrition and healthcare professionals, the findings emphasize the importance of considering KDs microbial effects and resulting health implications at an individual level.

**Keywords:** Ketogenic diet, Gut microbiota, Human.

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## Introduction

The ketogenic diet (KD), characterized by a macronutrient composition of low carbohydrates, high fat, and adequate protein (1), has traditionally been used to treat epilepsy (2), but evidence indicates its therapeutic use in other conditions, including cancer, neurodegenerative diseases (3), obesity, and type-2 diabetes (T2D) (4, 5). Additionally, athletes adopt KD to improve performance and reduce body fat (6).

KD may alter the gut's microbial composition that is fundamental to human health (7). As compositional imbalances are implicated in the development of certain diseases (8), it is important to determine the microbial changes induced by KD in order to understand these implications.

Existing reviews examining KD's impact on microbiota have collated evidence from murine and human studies (7, 9-12). However, differences between murine models and human systems limit their ability to recapitulate human microbial changes resulting from interventions, and caution is recommended in drawing conclusions about humans from murine studies (13). Furthermore, previous reviews have often included

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studies of infants. While research is not conclusive on when a child's microbiome reaches maturation, the consensus is the majority of development takes place before three years (14, 15). The exclusion of children under this age, as in this review, avoids bias as a result of developmental changes.

The current systematic review is novel in that it examines KD's impact on human gut microbiota in isolation, without the bias of potentially non-comparable murine models or human developing microbiomes. In another unique approach, this review then identifies potential consequent health outcomes specifically from these microbial impacts in order to make recommendations for practice and enable nutrition and healthcare professionals to make informed decisions when dealing with KD therapeutically.

### The ketogenic diet

KD is defined by its ability to induce a state of "physiological ketosis," with higher-than-normal ketone liver production (16, 17). Under normal dietary conditions, the majority of the body's tissues utilize glucose and fatty acids as energy sources, as both are metabolized to acetyl-coenzyme A, which condenses with oxaloacetate to enter the tricarboxylic acid cycle (18). During a KD, however, diminishing glucose blood levels and reserves instigate two processes to provide glucose and FAs for energy production; gluconeogenesis in the liver and lipolysis of FAs from adipose tissue (19). Consequently, the oxaloacetate supply becomes limited for two reasons. Firstly, glycolysis falls to low levels and oxaloacetate production relies on glycolysis generating its precursor pyruvate. Secondly, oxaloacetate is preferentially used in gluconeogenesis (20). Thus, the oxaloacetate supply becomes insufficient to condense all the acetyl-coenzyme A produced from FAs, and the liver diverts it to produce ketones as an alternative extra-hepatic energy source in the biochemical process "ketogenesis" (19, 20).

A KD may also be defined by the macronutrient ratios consumed. For the purposes of treating epilepsy, five categories have been defined: the classic KD, the modified KD, the medium-chain triglyceride (MCT) KD, the modified Atkins diet, and the low glycemic index treatment (LGIT) (1, 21). The ratio of fat types is specified in the MCT KD, and likewise, the ratios of

carbohydrate types is specified in the LGIT. However, there is much more variation in the macronutrient ratios seen in KD research and real life. It is accepted that ketosis results from restricting carbohydrates to under 50 grams (g), or 10% of total energy, while keeping protein adequate at 1.2–1.5 g per kilogram (kg) weight/day and making up the remaining energy intake percentage with fat, normally 60% to 90% (22, 23). Elevated serum levels of the ketone 3-beta-hydroxybutyrate ( $\beta$ OHB) indicate ketosis (20), with  $\beta$ OHB  $\geq$ 0.5 millimole (mmol)/litre (l) accepted as the threshold (24–26), although ketone production is subject to individual variability (27, 28) and influenced by fat and protein type (26, 29).

### Gut microbiota and health

The gut microbiota refers to the bacteria, archaea, and eukaryotes residing in the gastrointestinal tract (30). The majority colonize the colon, with bacterial numbers estimated at  $3.8 \times 10^{13}$ ; this review uses the term microbiota to refer specifically to these (31). Microbiota are predominantly from two phyla, *Bacteroidetes* and *Firmicutes*, while *Actinobacteria*, *Proteobacteria*, and *Verrucomicrobia* are present in smaller proportions (32). The principal genera comprise *Alistipes*, *Bacteroides*, *Faecalibacterium*, *Bifidobacterium*, *Eubacterium*, *Dorea*, and *Ruminococcus* (32). Despite the constancy of these constituents, dramatic differences are present among individuals in terms of relative proportions and species (33). It has been proposed that microbial composition can be categorized into three "enterotypes," namely *Bacteroides*, *Prevotella*, and *Ruminococcus*, distinguishable by the dense population of these genera (34). Microbiota population begins in utero and experiences volatile compositional shifts until around three years of age, when the diversity and composition resemble those of an adult (35, 36). Through adulthood, it is a relatively stable, core community of bacterial strains, with changes normally only in abundances (37, 38). Factors influencing composition include age, sex, geography (35, 39, 40), stress (41), drug use (42, 43), and diet (44, 45).

A symbiotic relationship exists between an individual and their microbiota. Microbiota are able to break down undigested macronutrients, producing an array of metabolites which, in turn, exhibit multifarious

effects on the host (46). Metabolites communicate locally and systemically through metabolic, immune, and neuroendocrine crosstalk with the host (47). Short-chain fatty acids (SCFAs) such as butyrate, propionate, and acetate are predominant metabolites produced mainly through carbohydrate fermentation by specific species (36, 48).

It is well recognized that microbiota are fundamental to human health, with their metabolites playing a key role in modulating disease risk (8, 49). Many metabolites, particularly SCFAs, are beneficial, supporting gastrointestinal integrity and immune system regulation (50, 51). Dysbiosis, where the microbiota's configuration adopts an abnormal state (52), is implicated in the development of cardiovascular disease, metabolic disorders, inflammatory bowel disease (IBD), and some cancers (49, 53).

### **Impact of KD on gut microbiota**

Diet is instrumental in shaping the microbiota's composition and activity (45, 54). Long-term dietary patterns are strongly associated with enterotype (55) and acute dietary interventions, as short as 24 hours, can instigate compositional changes, although it remains unknown what intervention length would translate to durable changes (56, 57). Diet type, for example, plant- versus animal-based, as well as specific dietary components, particularly macronutrients, have profound microbial impacts (8, 49, 56). Given KD's extreme alterations to dietary macronutrient ratios, it follows that this diet may instigate shifts in an individual's microbial composition.

Carbohydrates are commonly classified as either digestible or non-digestible carbohydrates (NDC). Reaching the colon in large quantities for fermentation, NDCs particularly influence microbiota, with variable and complex differences in impact depending on NDC type (8, 49, 58). Unsurprisingly, research thus indicates that a diet low in NDCs, such as KD, reduces bacterial abundances (9, 59).

Dietary fats are mainly absorbed in the small intestine, following emulsification by bile acids (58, 60). Small amounts reach the colon (61), with potentially potent antimicrobial properties (62, 63). Animal studies indicate a high-fat diet has a negative impact on microbiota composition (49, 63). However, a human study that administered a lipase inhibitor to increase fat reaching the colon demonstrated no

significant changes to microbial composition (61). Furthermore, the type of fat consumed appears to have differing microbial impacts (49, 63).

Protein is mainly digested and absorbed in the small intestine, yet factors such as source, processing, and macronutrient ratios affect its digestibility and the quantity and type of amino acids reaching the colon for bacterial fermentation, thus resulting in compositional changes (64, 65). Generally, consumption correlates with improved microbial diversity (49).

There are numerous other possible mechanisms by which KD influences metabolic and endocrine functions that may, in turn, impact gut microbiota (12).

## **Methods**

### **Research strategy**

The methods outlined by the Centre for Reviews and Dissemination (CRD) (2009) (66) guidance for undertaking systematic reviews in healthcare were followed in this study, which included quasi-experimental designs and RCTs, due to the limited amount of relevant research.

### **Data collection and search terms**

The Medical Literature Analysis and Retrieval System Online (MEDLINE), PubMed, and the Cumulative Index to Nursing and Allied Health Literature (CINAHL) were searched systematically (66, 67). The Cochrane Library was searched but did not offer any further relevant literature. Backward citation searches were performed on reference lists of selected studies (68). The terms used to locate literature relevant to the research objectives were "Low-carbohydrate" OR "low carbohydrate" OR "low carb" OR "carbohydrate restricted" OR "carbohydrate-restricted" OR ketogenic AND microbiome OR microbiota OR flora OR microbial.

### **Inclusion and exclusion criteria**

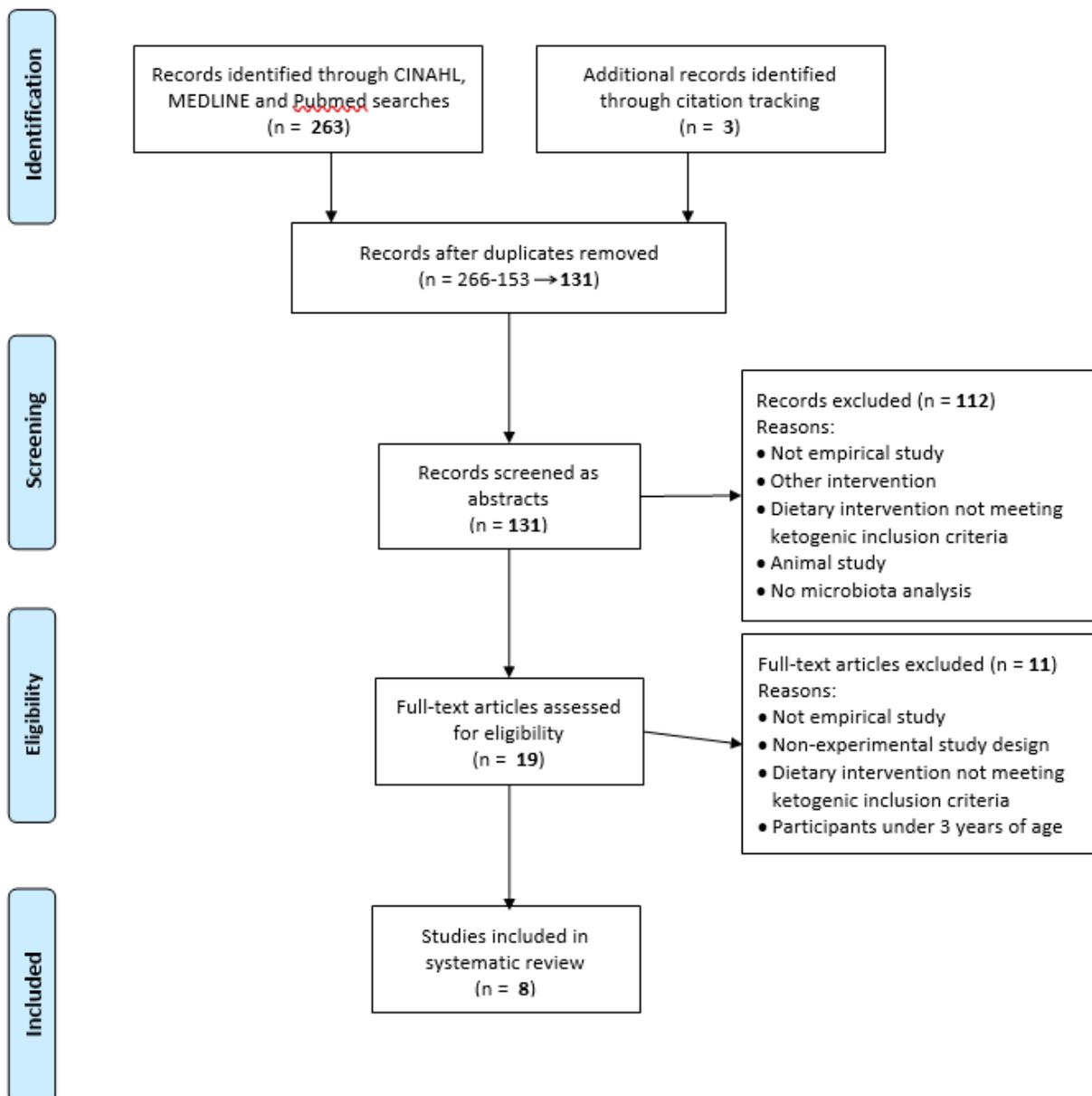
Table 1 details the inclusion and exclusion criteria of this review.

### **Study selection**

Figure 1 summarizes the study selection process using the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) flow diagram (69).

**Table 1.** Inclusion and exclusion criteria.

|               | Include  | Exclude   |
|---------------|--|---|
| Evidence type | Peer-reviewed empirical research, published in English   | Systematic/literature reviews<br>Meta-analyses<br>Grey literature |
| Population    | Humans, 3 years or older   | Children below 3 years<br>Animals or other organisms              |
| Intervention  | Ketosis demonstrated by blood $\beta$ OHB $\geq 0.5$ mmol/l and/or KD, with composition fat $\geq 60\%$ , carbs $\leq 50$ g/day or 10% energy and protein making up difference | KD not meeting the inclusion criteria<br>Any other intervention   |
| Comparator    | Any non-KD   | Diet that meets KD inclusion criteria                             |
| Outcomes      | Microbiota composition   | Microbiota composition unreported                                 |
| Study design  | RCTs<br>Quasi-experimental designs   | Non-experimental designs  |



**Figure 1.** Study selection flow diagram [Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA)].

## Appraisal and thematic analysis

The quality appraisal checklist developed by the National Institute of Care and Excellence (NICE) (2018) (70) for the development of public health guidance was adapted and utilized. No studies were excluded based on their quality because of the limited number of relevant papers and their possible value for investigating the research objectives (71). An adapted thematic analysis was undertaken to extract and collectively interpret relevant data from the complex information presented across the included studies (71, 72). To minimize errors, guidelines advise multiple researchers carry out the screening, selection, and data extraction (66, 73); this was not permitted for this Master's dissertation, however, the author built in checking processes.

## Results

### Study Characteristics

A summary of the characteristics of the eight included studies is presented in Table 2.

### Thematic analysis

The thematic analysis revealed seven themes, as detailed in Figure 2.

### Summary of results

Following their KD intervention, all included studies reported significant alterations to microbiota composition, and four studies, i.e. those investigating SCFAs, found disruptions to faecal SCFAs. This review focused on the phyla, genera, and species reported by multiple papers in order to synthesise and draw conclusions supported by a weight of empirical evidence (81). Table 3 displays a summary of the

**Table 2.** Summary of human studies investigating KD effects on faecal microbiota.

| Reference | Study Design                        | Population/Characteristics                     | N                                  | KD   | Intervention Length | Microbiota Analysis Method        |
|-----------|-------------------------------------|--|------------------------------------|--|---------------------|-----------------------------------|
| 74        | RCT – Parallel                      | Overweight/ obese<br>Age: 24–64<br>Sex: M & Fe | KD: 48<br>Control group: 43        | C 4%, F 61%, P 35%<br>Diet designed for weight loss, ≈ 30% energy restricted.      | 8-week              | Selective culture media           |
| 75        | RCT - Crossover                     | Obese<br>Age: 23–57<br>Sex: M                  | 18                                 | C 4%, F 66%, P 30%<br>Diet designed for weight loss, ad libitum intake.            | 4-week              | FISH                              |
| 76        | RCT - Crossover                     | Obese<br>Age: unreported<br>Sex: M             | 15                                 | C 4%, F 66%, P 30%<br>Designed for weight loss, ad libitum intake.                 | 4-week              | FISH                              |
| 77        | Experimental ITS                    | Obese & NAFLD<br>Age: 50-58<br>Sex: M & Fe     | 10                                 | C 4%, F 72%, P 24%<br>Diet designed to maintain weight.                            | 2-week              | Whole genome sequencing           |
| 78        | Non-randomised controlled trial     | Elite race walkers<br>Age: 20-35<br>Sex: M     | KD: 10<br>Control groups: 9 and 10 | C 5%, F 78%, P 17%<br>Diet designed to maintain weight.                            | 3-week              | 16S-rRNA gene amplicon sequencing |
| 79        | RCT - Crossover                     | Overweight/ obese<br>Age: 21-74<br>Sex: M      | 17                                 | C 5%, F 66%, P 29%<br>Diet designed for weight loss, fixed intake.                 | 4-week              | FISH                              |
| 80        | Experimental ITS                    | Multiple sclerosis<br>Age/Sex: Unreported      | 10                                 | C <50g, F >160g, P <100g to achieve βOHB ≥500 μmol/L (0.5 mmol/L)                  | 6-month             | FISH                              |
| 28        | Experimental before and after study | GLUT1-DS<br>Age: 8-34<br>Sex: M & Fe           | 6                                  | 1:1 ratio KD increased to 2:1, 3:1 or 4:1 as required to produce βOHB ≥2.0 mmol/l. | 3-month             | qPCR analysis                     |

16S-rRNA: 16S ribosomal ribonucleic acids; βOHB: beta-hydroxybutyrate; μmol/L; Micromoles per litre; C: Carbohydrate (as % energy); F: Fat (as % energy); Fe: Female; FISH: Fluorescence in situ hybridization; GLUT1-DS: Glucose transporter 1 deficiency syndrome; ITS: Interrupted time series; KD: Ketogenic diet; M: male; mmol/L: millimoles per litre; N: number of participants; NAFLD: Non-alcoholic fatty liver disease; P: Protein (as % energy); RCT: Randomised control trial; qPCR: real-time quantitative polymerase chain reaction

**Changes to microbiota composition, indicated by bacteria abundances**

- Theme 1 - Bacterial count and/or diversity alterations<sup>a</sup>
- Theme 2 - *Firmicutes* alterations<sup>a</sup>
- Theme 3 - *Bacteroidetes* alterations<sup>a</sup>
- Theme 4 - *Actinobacteria* alterations<sup>a</sup>

**Health implications of compositional changes**

- Theme 5 - SCFA alterations (key metabolites linking microbiota to health)<sup>a</sup>
- Theme 6 - Colonic health implications<sup>b</sup>
- Theme 7 - Health implications of reduced *Bifidobacterium*<sup>b</sup>

Notes: a. relevant results identified within included studies. b. relevant discussions/risks identified within included studies.

**Figure 2.** Themes identified through thematic analysis.

included studies’ results. Table 4 displays a summary of these results according to themes 1-5.

**Discussion**

**Mechanisms behind KD’s impact on gut microbiota**

This review indicates that KD has a general “anti-microbial” effect, reducing bacterial count, although potentially only in the short-term. Given the microbiota’s role in the breakdown of NDCs, research has predominantly allocated cause to the reduction in carbohydrate intake (82-84). Despite using inconsistent unitary measures of fibre, four studies (72, 74, 76, 79) examined NDC intake, and all confirmed intake on a KD is significantly reduced. NDC intake directly correlates with the amount of carbohydrate reaching the colon for bacterial fermentation (85).

This review’s most striking and conclusive finding relates to KD’s persistent negative impact on *Bifidobacterium*, which strictly metabolises carbohydrate substrates as its energy source (46, 86). Their central fermentation pathway, the fructose-6-phosphoketolase or “bifid shunt,” provides them with an ecological advantage in the presence of carbohydrates, because it produces more energy compared with the fermentation pathways of other bacterial species (87, 88). These insights may explain

its decreased abundance in response to the reduced NDC intake of a KD.

Furthermore, in addition to the liver, intestinal epithelial cells are able to produce ketones (89), and a KD results in increased  $\beta$ OHB within intestinal tissues (90). Recent in vitro and in vivo experiments have demonstrated that  $\beta$ OHB directly inhibits *Bifidobacterium* growth (91).

This review also provides reasonable evidence of a reduction in the butyrate-producing *Firmicutes*, *Eubacterium rectale*, *Roseburia*, and *Faecalibacterium prausnitzii*, although again, the results indicate this may not be a long-term concern due to potential microbial adaptation. *Eubacterium rectale*, *Roseburia*, and *Faecalibacterium prausnitzii* ferment NDCs directly. They also utilize *Bifidobacterium*’s fermentation metabolites in a process called substrate cross-feeding to produce butyrate (86, 92). Thus, a reduction in these bacteria could be explained by the concomitant decrease in both NDC intake and *Bifidobacterium*. Research has demonstrated high dietary fibre intake increases these bacteria (93). As more acidic conditions favor their growth, this is likely due to fermentation producing acidic end-products, such as SCFAs and lactic acid (94, 95). It follows that the fibre-restricting KD may have the opposite effect. Thus, there is a physiological basis for the KD causing a reduction in these bacteria.

### Colonic health implications

Colonic health was the main concern raised by the included studies relating to the KD's microbial impacts, primarily ascribed to the negative impact on SCFA

production (96). While this review may not provide conclusive evidence for this risk, the findings warrant further consideration.

SCFAs act through complex mechanisms to exert

**Table 3.** Summary of included study results relating to bacterial abundances and SCFAs.

| Study Reference | Themes 1-4: Changes to bacterial abundances  | Theme 5: SCFA alterations (faecal concentrations)   |
|-----------------|--|---|
| 74              | ↓: <i>Bifidobacterium</i> (p<0.001) in KD group but not in control group.<br>No significant change: <i>Lactobacilli</i> for either KD or control groups.   | ↓: Total SCFAs, acetate, butyrate (p≤0.04), in KD group but not for control group.  |
| 75              | ↓: total bacterial count (p<0.001), <i>Roseburia</i> and <i>Eubacterium rectale</i> (p<0.001), and <i>Bifidobacterium</i> (p<0.026) for both medium carbohydrate control and KD groups, with a progressive gradient as dietary carbohydrate reduced.<br><br>No significant change: <i>Bacteroides</i> , <i>Faecalibacterium prausnitzii</i> , <i>Firmicutes</i> , <i>Lactobacillus-Enterococcus</i> group and <i>Desulfovibrio</i> genus for either medium carbohydrate control or KD groups.  | ↓: Total SCFAs, acetate, butyrate, isovalerate, propionate, valerate (p<0.05) for both medium carbohydrate control and KD groups.<br>Butyrate reductions showed significant progressive gradient as dietary carbohydrate reduced. |
| 76              | Note: Results combined with 70. Duncan <i>et al.</i> (2007).<br>↓: Total bacterial count (p<0.001), <i>Roseburia</i> and <i>Eubacterium rectale</i> (p<0.001), and <i>Bifidobacterium</i> (p<0.037).<br>No significant change: <i>Bacteroides</i> , <i>Faecalibacterium prausnitzii</i> , <i>Firmicutes</i> .  |   |
| 77              | Note: Significant changes to 94 bacterial strains across 25 genera. 10 most abundant genera reported here.<br>↓: <i>Ruminococcus</i> (p=2.62e-09), <i>Eubacterium</i> (p=2.84e-08), <i>Clostridium</i> (p=3.73e-12), <i>Coprococcus</i> (p=0.0056), <i>Bifidobacterium</i> (p=6.77e-14), <i>Subdoligranulum</i> (p=0.00039), <i>Butyrivibrio</i> (p=1.65e-05).<br>↑: <i>Streptococcus</i> (p=0.0014), <i>Lactococcus</i> (p=2.32e-05), <i>Eggerthella</i> (p=0.0073).  | ↓: Total SCFAs (p=0.047).   |
| 78              | ↓: <i>Faecalibacterium</i> (p=0.007) <i>Bifidobacterium</i> , <i>Veillonella</i> , <i>Streptococcus</i> , <i>Succinivibrio</i> , <i>Odoribacter</i> and <i>Lachnospira</i> (no p-values provided as identified through LefSe and/or sPLS-DA).<br>↑: <i>Dorea</i> (p=0.007), <i>Bacteroides</i> (p=0.002), <i>Enterobacteriaceae</i> , <i>Peptostreptococcaceae</i> , <i>Barnesiellaceae</i> and <i>Akkermansia</i> (no p-values provided as identified through LefSe and/or sPLS-DA).<br>No significant change: Alpha-diversity.   |   |
| 79              | ↓: Total bacteria count (p=0.013) and <i>Bacteroides</i> (P=0.007) for both medium carbohydrate control and KD groups, <i>Roseburia</i> and <i>Eubacterium rectale</i> (P<0.001) for only KD group.<br>No significant change: <i>Lachnospiraceae</i> or <i>F. prausnitzii</i> .  | ↓: Total SCFAs, acetate, butyrate (p<0.001) in KD group only.<br>↓: Isobutyrate (p=0.002), isovalerate (p<0.001) and propionate (p<0.001) for both medium carbohydrate control and KD groups.                                     |
| 80              | At 2 weeks<br>↓: Diversity (p=0.03-0.05), total bacteria count (p<0.001), <i>Bacteroides</i> (p=0.001-0.002), <i>Faecalibacterium prausnitzii</i> (p=0.001), <i>Bifidobacterium</i> (p=0.02-0.03), <i>Atopobium</i> cluster (p=0.02), <i>Ruminococcus albus</i> (p=0.02) and <i>Sphaerotilus natans</i> (p=0.02).<br>At 6 months<br>↓: <i>Bifidobacterium</i> (p<0.001), <i>Coriobacterium</i> (p=0.02).<br>↑: Total bacteria count (p=0.02), <i>Roseburia</i> and <i>Eubacterium rectale</i> (p=0.03-0.02), <i>Clostridium viride</i> (p<0.01), <i>Eubacterium hallii</i> (p<0.01), <i>Ruminococcus productus</i> (p=0.03).<br>No significant change: Diversity or the other 29 investigated bacterial species. |   |
| 28              | ↑: <i>Desulfovibrio</i> (p=0.025).<br>No significant change: <i>Firmicutes</i> , <i>Bacteroidetes</i> , <i>Lactobacillus</i> , <i>Bifidobacterium</i> , <i>Faecalibacterium prausnitzii</i> , <i>Clostridium perfringens</i> , <i>Enterobacteriaceae</i> and <i>Desulfovibrio</i> .  |   |

↑: increased abundance; ↓: decreased abundance; LefSe: Linear discriminant analysis effect size; sPLS-DA: Sparse partial least squares discriminant analysis; SCFA: Short chain fatty acid.

beneficial effects in the colon (97). Butyrate, the primary energy source for colonocytes, supports intestinal barrier

structure and function and protects from external harm by facilitating epithelial tight-junction assembly, stimulating

**Table 4.** Summary of results for themes 1-5.

| Theme                                     | Summary of results  |
|---|---|
| Theme 1: Bacterial count and/or diversity | <p>Total bacterial count:</p> <ul style="list-style-type: none"> <li>Highly significant evidence from four studies of a reduction in total bacterial numbers with KD adherence between 2-12 weeks. [75 (p&lt;0.001), 76 (p=0.013), 79 (p&lt;0.001), 80 (p&lt;0.001)].</li> <li>One study indicates longer-term adherence may reverse effect, reporting increased bacterial numbers after 6 months [80 (p=0.02)].</li> </ul> <p>Bacterial diversity:</p> <ul style="list-style-type: none"> <li>Inconsistent diversity data from two studies provides insufficient evidence of KD effects [80 reported a reduction in diversity (p=0.03-0.05) at 2-12 weeks but this was reversed to baseline levels by 6 months. 78 reported no change in alpha-diversity, or richness].</li> </ul>   |
| Theme 2: Firmicutes                       | <p><i>Firmicutes</i> phylum:</p> <ul style="list-style-type: none"> <li>Three studies found no significant changes to <i>Firmicutes</i> abundance following a KD. [75;76;28]</li> </ul> <p><i>Lactobacillus</i> genus:</p> <ul style="list-style-type: none"> <li>Four studies found no significant changes to <i>Lactobacillus</i> abundances following a KD [75;74;80;28].</li> </ul> <p><i>Eubacterium</i> genus, <i>Eubacterium rectale</i> and <i>Roseburia</i>:</p> <ul style="list-style-type: none"> <li>One study found highly significant reductions of <i>Eubacterium</i> genus [77 (p=2.84e-08)] and of <i>Eubacterium rectale</i> species [77 (p=4.91e-25)], with KD adherence.</li> <li>Additional significant evidence from three studies for the reduction of <i>Eubacterium rectale</i> and <i>Roseburia</i> (collectively) following KD [75;76;79 (p&lt;0.001)]</li> <li>Conflicting evidence from one study, reporting no change to <i>Eubacterium rectale</i> and <i>Roseburia</i> abundance at 2 and 12 weeks of KD adherence and a slight increase by 6 months [80 (p=0.03-0.02 compared with baseline, 2 and 12 weeks)].</li> </ul> <p><i>Faecalibacterium prausnitzii</i>:</p> <ul style="list-style-type: none"> <li>Significant findings for the reduction of <i>Faecalibacterium prausnitzii</i> abundance following KD adherence of 2-3 weeks from three studies [77 (p=0.000), 78 (p=0.007), 80 (p=0.001)].</li> <li>One study indicates longer-term adherence may reverse effect, reporting reversal to baseline levels after 6 months KD adherence [80].</li> <li>Conflicting evidence from three studies reporting no significant change in <i>Faecalibacterium prausnitzii</i> abundance following their KD interventions [75;79;28].</li> </ul> |
| Theme 3: Bacteroidetes                    | <p><i>Bacteroidetes</i> phylum:</p> <ul style="list-style-type: none"> <li>Only one study reported no significant change to <i>Bacteroidetes</i> phylum as a result of their KD intervention [28].</li> </ul> <p><i>Bacteroides</i> genus:</p> <ul style="list-style-type: none"> <li>One study reported a significant increase in <i>Bacteroides</i> [78 (p=0.002)].</li> <li>Two studies reported no significant change to <i>Bacteroides</i> [75;76].</li> <li>Conversely, two studies observed a decrease in <i>Bacteroides</i> with KD adherence of between 2-12 weeks [79 (p=0.007), 80 (p=0.002-0.001)]</li> <li>One study indicates longer-term adherence may reverse any decrease, reporting reversal to baseline levels after 6 months KD adherence [80]</li> </ul>   |
| Theme 4: Actinobacteria                   | <p><i>Bifidobacterium</i> genus:</p> <ul style="list-style-type: none"> <li>Five studies provide highly significant data demonstrating reduced <i>Bifidobacterium</i> resulting from short-term and longer-term KD adherence [74 (p&lt;0.001), 75 (p&lt;0.001), 76 (p&lt;0.001), 77 (p=6.77e-14), 80 (p=0.02-0.03 at 2 and 12 weeks, p&lt;0.001 at 6 months)].</li> <li>One study detected a reduction using Linear discriminant analysis effect size (LefSe) and Sparse partial least squares discriminant analysis (sPLS-DA), but not in their initial multivariate statistical analysis methods, Redundancy Analysis or Anosim [78].</li> <li>One study observed no change in <i>Bifidobacterium</i> abundance [28].</li> </ul>  |
| Theme 5: SCFAs                            | <p>Total faecal SCFAs:</p> <ul style="list-style-type: none"> <li>Supporting evidence from four studies for a reduction in total faecal SCFAs in response to a KD [74 (p≤0.04), 75 (p&lt;0.05), 76 (p=0.047), 77 (p&lt;0.001)].</li> </ul> <p>Acetate and butyrate (faecal):</p> <ul style="list-style-type: none"> <li>Three studies demonstrated significant reductions in acetate and butyrate [74 (p≤0.04), 75 (p&lt;0.05) and-79].</li> </ul> <p>Propionate (faecal):</p> <ul style="list-style-type: none"> <li>Two studies demonstrated significant reductions in propionate [75 (p&lt;0.05), 79 (p=0.047)].</li> <li>One study found no significant reduction to propionate [74].</li> </ul>  |

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mucin production, and inhibiting pathogenic bacterial adhesion (99). SCFAs act as the microbiota's link with the host's immune system through several cellular signaling pathways that ultimately modify processes such as gene expression, differentiation, proliferation, and apoptosis (100). They activate G-coupled protein receptors (GPRs), which are expressed in the colonic mucosa and link to downstream signaling pathways involved in gut immune homeostasis (97, 100). They constrain the enzymatic actions of histone deacetylases (HDACs) in colonocytes and mucosal immune cells, which inhibits DNA transcription and the regulation of inflammatory-associated gene expression (97, 100). Through these actions, SCFAs play a preventative role in the development of IBD, Crohn's disease, and ulcerative colitis (97, 101). Reduced faecal butyrate levels have been detected in IBD patients and ulcerative colitis patients in remission compared with controls (102, 103). SCFAs' aforementioned actions protect against colorectal cancer, and they induce apoptosis and inhibit proliferation of colonic cancer cells (97, 101).

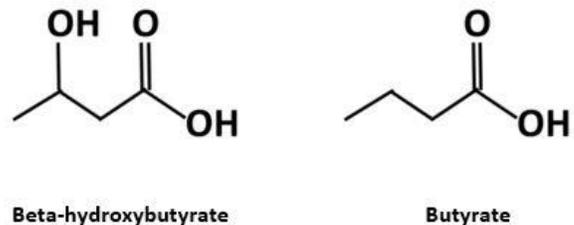
Microbiota are considered key in the pathogenesis of IBD and colorectal cancer (104-106) with dysbiotic compositions identified in patients, characterized by reduced butyrate-producing *Firmicutes*, especially *Faecalibacterium prausnitzii* in IBD patients (107) and *Roseburia* in colorectal cancer patients (108).

High *Bifidobacterium* abundance is important for colonic health (109), intestinal barrier function (110), and facilitating healthy microbial composition through cross-feeding metabolites to butyrate-producers and competitive exclusion of pathogenic bacteria (109, 111). Their metabolites have direct benefits, with certain strains prolifically producing conjugated linoleic acid (112), which is anti-inflammatory and inhibits colon cancer cell growth and proliferation (96). Murine models have demonstrated that *Bifidobacterium* provides protection from certain carcinogens (103) and has anti-tumor effects (114). Furthermore, reduced *Bifidobacterium* abundance is found in colorectal cancer patients (115).

Cumulatively, this research suggests that the pattern of microbial alterations following KD adherence, namely decreased *Bifidobacterium* and *Firmicutes* butyrate-producing bacteria, may be detrimental to colonic health and potentially increase the risk of colonic diseases. However, to the best of the authors

knowledge, no studies have investigated these risks specifically for KD. There is extensive research into the relationship between diet, microbiota, SCFA alterations, and colonic diseases, with fibre a key focus, given it promotes *Bifidobacterium* and *Firmicutes* abundance as well as SCFA production. However, there remains controversy on its criticality for colonic health; epidemiological studies provide abundant evidence of an inverse association between fibre intake and colorectal cancer risk, yet other cohort studies have found no association (106, 116), and RCTs investigating the effect of supplementation for the prevention of colorectal cancer have yielded inconsistent results (117). A recent study found no association between faecal SCFA concentrations and colonic carcinomas (118). For IBD, the consensus is that fibre reduces the risk of disease development (119), although the beneficial effect of a high-fibre diet or supplementation for IBD patients has not been demonstrated (120). Inconsistency across such research may not be surprising, given fibre is not a homogeneous substance, rather a group of compounds with differing properties and effects on the microbiota and SCFA production, with type, dose, and consumption timing all likely influencing outcomes (120, 121).

For KD, there is a further consideration relating to the fact that ketone,  $\beta$ OHB, has a similar chemical structure to butyrate, as depicted in Figure 3.



**Figure 3.** Chemical structures of beta-hydroxybutyrate and butyrate.

Consequently, they have functional similarities, for example,  $\beta$ OHB activates GPRs, such as GPR109a (122, 123), and inhibits HDACs (124, 125). It has been hypothesized that the increased systemic levels of  $\beta$ OHB induced during KD may lessen the importance of microbial butyrate production (126), and thus, the potential negative impact on colonic health from reduced butyrate may be lessened.

## Health implications of reduced *Bifidobacterium*

*Bifidobacterium*'s metabolic activities are considered fundamental to human health (96), and several extraintestinal diseases have been associated with reduced *Bifidobacterium*, namely obesity, T2D (128-131), and depression (132). While currently no research provides definitive evidence of a causal relationship, wider research does provide corroboration and probable physiological mechanisms.

For obesity and T2D, SCFA production and metabolic endotoxemia are two mechanisms postulated to link altered microbiota compositions with obesity and metabolic syndrome (133). *Bifidobacterium* mediates the production of SCFAs, which influence signaling pathways through GPR activation. These actions extend beyond the gut to stimulate production of several appetite regulating hormones, including adipocyte-derived leptin (134). Leptin acts on the hypothalamus to suppress appetite and interacts with insulin signaling (135). SCFAs stimulate two intestinal-derived hormones, glucagon-like peptide and peptide YY (136, 137), which play important roles in glucose homeostasis and promoting satiety (138, 139). Metabolic endotoxemia is defined as an increase in plasma bacterial lipopolysaccharide (LPS) (140). In mice, the link between high-fat diet-induced microbial alterations, including a reduction in *Bifidobacterium*, and a downstream inflammatory response, specifically cytokines linked to insulin resistance, has been demonstrated (140, 141). In humans, high fat consumption has been demonstrated to induce metabolic endotoxemia (142), which in turn induces adipose inflammation and insulin resistance (143) and is associated with increased energy intake (144). LPS plasma levels rise as a result of increased intestinal permeability, allowing the flow of gram-negative bacteria, such as *Bacteroidetes* whose membranes contain LPS, from the intestines into the blood (145, 146). Many factors may contribute to a disrupted intestinal barrier function, but reduced *Bifidobacterium* may be key, given its role in preserving mucosal health. Thus, theoretically, a reduction in *Bifidobacterium* could promote obesity and T2D by instigating hormonal and immune disturbances. This is supported by murine trials, which demonstrate that *Bifidobacterium* probiotics lower plasma LPS (146)

and decrease body weight in mice fed a high-fat diet and have a wide range of other anti-obesity effects, including improved glucose homeostasis and decreased serum leptin (147, 148). Recent murine research, however, has highlighted that KD is distinctive in its impact on gut microbiota because of the production of ketone bodies which directly inhibit *Bifidobacterium* growth and, in turn, decreases the pro-inflammatory Th17 cells within the small intestine and possibly also adipose tissues (91). While it is postulated that this may be a possible mechanism in the reduction of body fat associated with KD (77, 91), whether this impact of *Bifidobacterium* on Th17 cells is beneficial or detrimental to gut health and inflammatory-related diseases is dependent on context and requires further investigation (149).

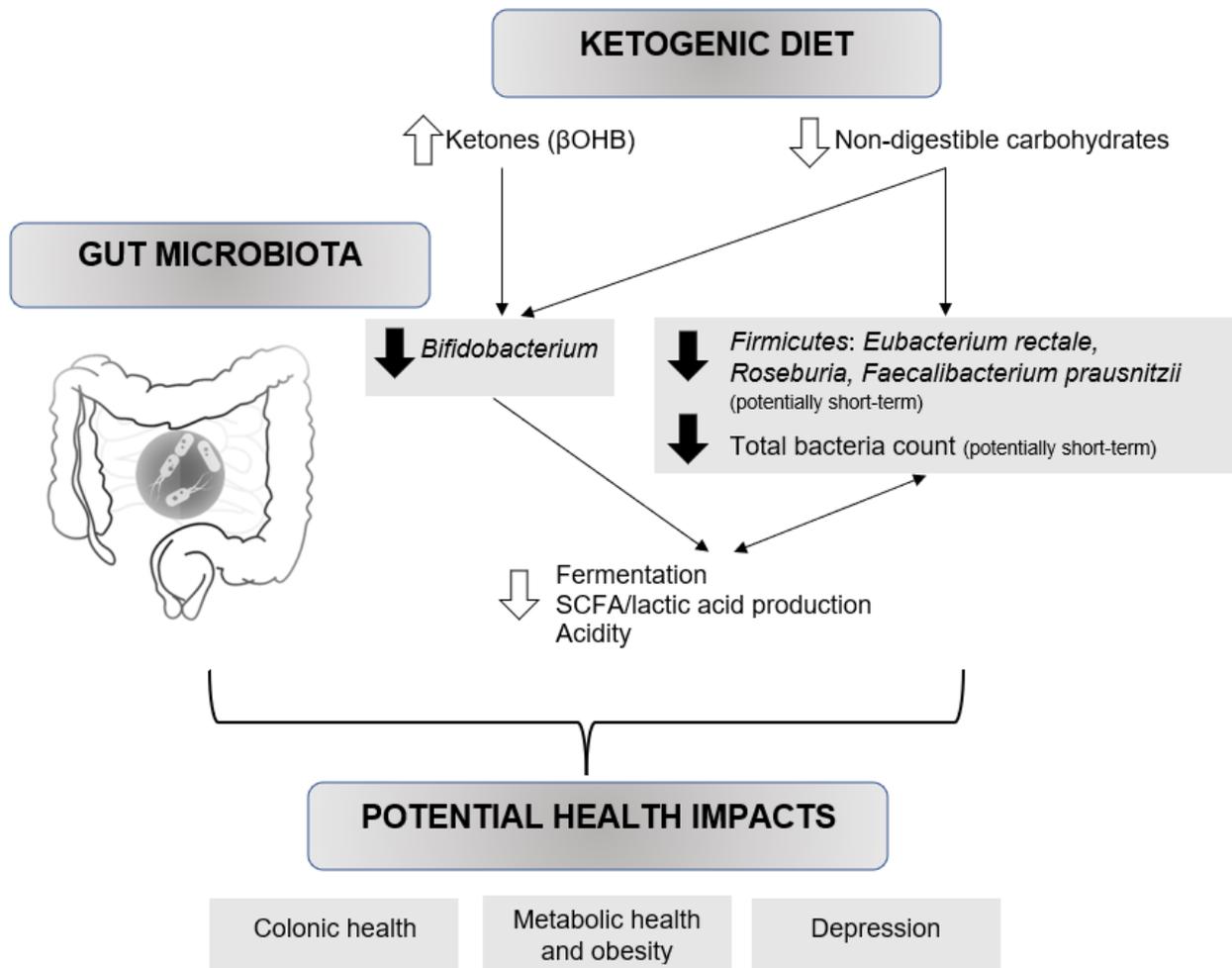
Microbiota are considered key in determining the onset and duration of depression through a complex array of mechanisms (150). Changes in microbiota composition alter the balance of chemicals produced, which in turn impacts the bi-directional communication between endocrine, immune, and central nervous systems, or the "gut-brain-axis" (151). The vagal nerve is the primary communication pathway between the microbiota and the brain, and its afferent receptors are stimulated by SCFAs and LPS (152). As discussed above, these bacterial metabolites are sensitive to *Bifidobacterium* abundance. Depression has been linked to an immune response instigated by increased intestinal permeability and the resulting raised serum LPS (145, 150). Disrupted brain neurotransmitters have been implicated in depression, including both gamma-aminobutyric acid (GABA) and serotonergic systems (151, 153). *Bifidobacterium* is a prolific GABA producer (154). While the ability of such microbiota-produced neurotransmitters to cross the blood-brain barrier is unclear (155), they do influence the central nervous system via the vagal nerve and its afferent receptors (156). Indeed, the administration of *Bifidobacterium longum* in mice displaying anxiety behaviors had anti-anxiolytic effects, which were demonstrated to be dependent on vagal nerve integrity (157). Microbiota modulate the metabolism of serotonin's precursor tryptophan to influence brain levels (150), as demonstrated in a study investigating the anti-depressive effects of *Bifidobacterium infantis* on rats, where administration resulted in reduced

depressive behaviors alongside increased plasma tryptophan (158). Additionally, a human RCT with irritable bowel syndrome patients demonstrated that *Bifidobacterium longum* altered brain activity and reduced depression scores (159). Thus, although the potential preventative effects of a KD on depression via its nutrition and microbiome impacts continue to attract attention (12), it is conceivable that a persistent disruption to *Bifidobacterium* abundance, as a result of longer-term KD adherence, could increase the risk of developing or worsening depression. Summary of findings is in Figure 4.

**Research strengths and limitations**

The key strengths of the current review are its systematic approach and its exclusive examination of human studies, excluding animal results that may not reflect human microbial responses. The inclusion of differing study designs ensures consideration of all

valuable evidence. However, this heterogeneity may hamper comparability and the ability to determine conclusive outcomes, as may the small number of included studies with mostly low participant numbers. The included studies lack consistency across factors that may influence microbiota, such as their KD composition, data collection methods, microbial analysis methods, inclusion and exclusion criteria, along with participant characteristics such as age, health, medication, and baseline diet. Furthermore, no included study demonstrated sufficient power, despite microbiome-specific power analysis approaches being available (160, 161). Microbiota-host interactions are complex and the health effects are not fully understood, limiting the health impact interpretation of microbial changes. KD’s effect on mucosal microbiota levels cannot conclusively be determined, as the included studies utilized faecal microbiota as a proxy.



**Figure 4.** Summary of findings

### The ketogenic diet in practice: Key messages for nutrition and healthcare professionals

- Consider faecal testing to assess microbiota composition and ascertain an individual's risks and priorities for microbial support during and/or post-KD adherence. An understanding of the commercial tests available, their interpretation and limitations is essential (163).
- Consider preserving or enhancing microbial communities, for example by:
  - Promoting NDC-containing foods (within constraints of KD, as required) or by supplementation (164).
  - Promoting dietary or lifestyle factors to support a healthy microbiota, such as exercise (165)
  - Discouraging dietary or lifestyle factors with potential negative microbial impacts, such as low-calorie sweeteners (166).
  - Probiotic supplementation may beneficially impact the microbiota's composition and metabolism, albeit transitorily (167;168).
- Tailor approach to individual health, disease status and purpose for KD adherence:
  - For individuals undertaking KD for weight loss, to manage T2D or with a history of depression, *Bifidobacterium* preservation should be a key priority.

### Conclusion

This review reveals that in humans, certain bacterial abundances and their metabolites are disrupted by KD adherence, most conclusively a decrease in *Bifidobacterium* and faecal SCFAs as well as short-term reductions in *Firmicutes* butyrate-producing bacteria and that these effects are possibly detrimental to colonic health. The persistent reduction in *Bifidobacterium* abundance may have additional detrimental impacts, with obesity, T2D, and depression as key risks.

### Recommendations for future research

Microbiome research is an evolving field, and development is required in key areas, such as cross-study comparability issues, particularly technical variation in sampling and microbial analysis of the observed microbial communities, their structure, and the biological conclusions drawn (162), as well as heterogeneity between participant baseline microbiota compositions (58).

To facilitate cross-study comparison and enable cause-effect conclusions in research into KD's impact, larger-scale, longer-term RCTs in healthy and specific population groups are required, with standardization of KD's composition and controlling of multifarious confounders.

Further research is required to confirm the complex interactions and causal relationships between microbes, their metabolites, and humans. For KD, research is warranted into the biological effects of  $\beta$ OHB and whether these might negate health impacts relating to reduced microbial butyrate production.

### Conflict of interests

The authors declare that there is no conflict of interests with respect to the research, authorship or publication of this article.

### References

1. Miranda MJ, Turner Z, Magrath G. Alternative diets to the classical ketogenic diet-Can we be more liberal? *Epilepsy Res* 2012;100:278–285.
2. Wheless JW. History of the ketogenic diet. *Epilepsia* 2008;49:3–5.
3. Branco AF, Ferreira A, Simões RF, Magalhães-Novais S, Zehowski C, Cope E, et al. Ketogenic diets: From cancer to mitochondrial diseases and beyond. *Eur J Clin Invest* 2016;46:285–298.
4. Kalra S, Singla R, Rosha R, Dhawan M. Ketogenic diet: Situational analysis of current nutrition guidelines. *J Pak Med Ass* 2018;68, pp. 1836–1839.
5. Bolla AM, Caretto A, Laurenzi A, Scavini M, Piemonti L. Low-carb and ketogenic diets in type 1 and type 2 diabetes. *Nutrients* 2019;11:962.
6. Gregory RM, Hamdan H, Torisky DM, Akers JD. A low-carbohydrate ketogenic diet combined with 6-weeks of crossfit training improves body composition and performance. *Int J Sports Ex Med* 2017;3:054.
7. Paoli A, Mancin L, Thomas E, Mota JF, Piccini F. Ketogenic diet and microbiota: Friends or enemies?. *Genes* 2019;10:534.
8. Mills S, Stanton C, Lane JA, Smith GJ, Ross RP. Precision nutrition and the microbiome, part I: Current state of the science. *Nutrients* 2019;11:1–45.
9. 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;11:373.
10. Dowis K, Banga S. The potential health benefits of the ketogenic diet: a narrative review. *Nutrients* 2021;13:1654.
11. Attaye I, van Oppenraaij S, Warmbrunn MV, Nieuwdorp M. The role of the gut microbiota on the beneficial effects of ketogenic diets. *Nutrients* 2022;14:191.

12. Zhu H, Bi D, Zhang Y, Kong C, Du J, Wu X, et al. Ketogenic diet for human diseases: the underlying mechanisms and potential for clinical implementations. *Signal Transduct Target Ther* 2022;7:11.
13. Nguyen TLA, Vieira-Silva S, Liston A, Raes J. How informative is the mouse for human gut microbiota research?. *Dis Models Mech* 2015;8:1–16.
14. Yatsunenko T, Rey F, Manary M, Trehan I, Dominguez-Bello M., Contreras M, et al. Human gut microbiome viewed across age and geography. *Nature* 2012;486:222–227.
15. Rodríguez JM, Murphy K, Stanton C, Ross RP, Kober OI, Juge N, et al. The composition of the gut microbiota throughout life, with an emphasis on early life. *Microb Ecol Health Dis* 2015;26:26050.
16. Krebs HA. The regulation of the release of ketone bodies by the liver. *Adv Enzyme Reg* 1966;4:339–354.
17. Cahill GF. Fuel Metabolism in Starvation. *Annu Rev Nutr* 2006;26:1–22.
18. El Bacha T, Luz M, Da Poian A. Dynamic Adaptation of Nutrient Utilization in Humans. *Nat Educ* 2010;3:8.
19. Clifton P, Brouns F. Low-carbohydrate diets: Nutritional and physiological aspects. *Obes Rev* 2006;7:49–58.
20. Laffel L. Ketone bodies: a review of physiology, pathophysiology and application of monitoring to diabetes. *Diabetes/Metab Res Rev* 1999;15:412–426.
21. Liu YM, Wang HS. Medium-chain triglyceride ketogenic diet, an effective treatment for drug-resistant epilepsy and a comparison with other ketogenic diets. *Biomed J* 2013;36:9–15.
22. Westman EC, Feinman RD, Mavropoulos JC, Vernon MC, Volek JS, Wortman JA, et al. Low-carbohydrate nutrition and metabolism. *Am J Clin Nutr* 2007;86:276–284.
23. Paoli A. Ketogenic diet for obesity: Friend or foe?. *Int J Environ Res Public Health*. 2014;11:2092–2107.
24. Guerci B, Benichou M, Floriot M, Bohme P, Fougnot S, Franck P, et al. Accuracy of an electrochemical sensor for measuring capillary blood ketones by fingerstick samples during metabolic deterioration after continuous subcutaneous insulin infusion interruption in type 1 diabetic patients. *Diabetes Care* 2003;26:1137–1141.
25. Volek JS, Phinney SD. The art and science of low carbohydrate living. 2011; 1st edn. Lexington: Beyond Obesity LLC.
26. Harvey CJDC, Schofield GM, Williden M. The use of nutritional supplements to induce ketosis and reduce symptoms associated with keto-induction: a narrative review. *Peer J* 2018;6:4488.
27. Schoeler NE, Cross JH. Ketogenic dietary therapies in adults with epilepsy: a practical guide. *Pract Neurol* 2016;1:208–214.
28. Tagliabue A, Ferraris C, Uggeri F, Trentani C, Bertoli S, de Giorgis V, et al. Short-term impact of a classical ketogenic diet on gut microbiota in GLUT1 Deficiency Syndrome: a 3-month prospective observational study. *Clin Nutr ESPEN* 2017;17:33–37.
29. Westerterp-Plantenga MS, Lemmens SG, Westerterp KR. Dietary protein - Its role in satiety, energetics, weight loss and health. *Br J Nutr* 2012;108:105–112.
30. Marchesi JR, Ravel J. The vocabulary of microbiome research: A proposal. *Microbiome* 2015;3:31.
31. Sender R, Fuchs S, Milo R. Revised estimates for the number of human and bacteria cells in the body. *PLOS Biol* 2016;14:1002533.
32. Tap J, Mondot S, Levenez F, Pelletier E, Caron C, Furet JP, et al. Towards the human intestinal microbiota phylogenetic core. *Environ Microbiol* 2009;11:2574–2584.
33. Lozupone C, Stombaugh J, Gordon J, Jansson J, Knight R. Diversity, stability and resilience of the human gut microbiota. *Nature* 2012;489:220–230.
34. Arumugam M, Raes J, Pelletier E, Le Paslier D, Yamada T, Mende DR, et al. Enterotypes of the human gut microbiome. *Nature* 2011;473:174–180.
35. Faith JJ, Guruge JL, Charbonneau M, Subramanian S, Seedorf H, Goodman AL, et al. The long-term stability of the human gut microbiota. *Science* 2013;341:1–22.
36. Rajilić-Stojanović M, Heilig HGHJ, Tims S, Zoetendal EG, De Vos WM. Long-term monitoring of the human intestinal microbiota composition. *Environ Microbiol* 2013;15:1146–1159.
37. Mueller S, Saunier K, Hanisch C, Norin E, Alm L, Midtvedt T, et al. Differences in faecal microbiota in different European study populations in relation to age, gender, and country: a cross-sectional study. *Appl Environ Microbiol* 2006;72:1027–1033.
38. Kim YS, Unno T, Kim BY, Park MS, et al. Sex differences in gut microbiota. *World J Men's Health* 2019;37:1–13.
39. Karl PJ, Hatch AM, Arcidiacono SM, Pearce SC, Pantoja-Feliciano IG, Doherty LA, et al. Effects of psychological, environmental and physical stressors on the gut microbiota. *Front Microbiol* 2018;9:1–32.
40. Iizumi T, Battaglia T, Ruiz V, Perez Perez GI. Gut microbiome and antibiotics. *Arch Med Res* 2017;48:727–734.
41. Maier L, Pruteanu M, Kuhn M, Zeller G, Telzerow A, Anderson EE, et al. Extensive impact of non-antibiotic drugs on human gut bacteria. *Nature* 2018;555:623–628.
42. Walker AW, Ince J, Duncan SH, Webster LM, Holtrop G, Ze X, et al. Dominant and diet-responsive groups of bacteria within the human colonic microbiota. *ISME J* 2011;5:220–230.
43. Conlon MA, Bird AR. The impact of diet and lifestyle on gut microbiota and human health. *Nutrients* 2015;7:17–44.
44. Oliphant K, Allen-Vercoe E. Macronutrient metabolism by the human gut microbiome: Major fermentation by-products and their impact on host health. *Microbiome* 2019;7:91.
45. Kho ZY, Lal SK. The human gut microbiome - a potential controller of wellness and disease. *Front Microbiol* 2018;9:1835.
46. Koh A, De Vadder F, Kovatcheva-Datchary P, Bäckhed F. From dietary fibre to host physiology: short-chain fatty acids as key bacterial metabolites. *Cell* 2016;165:1332–1345.
47. Singh RK, Chang HW, Yan D, Lee KM, Ucmak D, Wong K, et al. Influence of diet on the gut microbiome and implications for human health. *J Transl Med* 2017;15:73.
48. Morrison DJ, Preston T. Formation of short chain fatty acids by the gut microbiota and their impact on human metabolism. *Gut Microbes* 2016;7:189–200.

49. Lin L, Zhang J. Role of intestinal microbiota and metabolites on gut homeostasis and human diseases. *BMC Immunol* 2017;18:1–25.
50. Levy M, Kolodziejczyk AA, Thaïss CA, Elinav E. Dysbiosis and the immune system. *Nat Rev Immunol* 2017;17:219–232.
51. Marchesi JR, Adams DH, Fava F, Hermes GDA, Hirschfield GM., Hold G, et al. The gut microbiota and host health: a new clinical frontier. *Gut* 2016;65:330–339.
52. Graf D, Di Cagno R, Fåk F, Flint HJ, Nyman M, Saarela M, et al. Contribution of diet to the composition of the human gut microbiota. *Microb Ecol Health Dis* 2015;26:26164.
53. Wu GD, Chen J, Hoffmann C, Bittinger K, Chen YY, Keilbaugh SA, et al. Linking long-term dietary patterns with gut microbial enterotypes. *Science* 2011;334:105–108.
54. 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;505:559–563.
55. Klimentenko NS, Tyakht AV, Popenko AS, Vasiliev AS, Altukhov IA, Ischenko DS, et al. Microbiome responses to an uncontrolled short-term diet intervention in the frame of the citizen science project. *Nutrients* 2018;10:576.
56. Portune KJ, Benítez-Páez A, Del Pulgar EMG, Cerrudo V, Sanz Y. Gut microbiota, diet, and obesity-related disorders—The good, the bad, and the future challenges. *Mole Nutr and Food Res* 2017;61:1–17.
57. Chen HM, Yu YN, Wang JL, Lin YW, Kong X, Yang CQ, et al. Decreased dietary fibre intake and structural alteration of gut microbiota in patients with advanced colorectal adenoma. *Am J Clin Nutr* 2013;97:1044–1052.
58. Scott KP, Gratz SW, Sheridan PO, Flint HJ, Duncan SH. The influence of diet on the gut microbiota. *Pharmacol Res* 2013;69:52–60.
59. Morales P, Fujio S, Navarrete P, Ugalde JA, Magne F, Carrasco-Pozo C, et al. Impact of dietary lipids on colonic function and microbiota: an experimental approach involving orlistat-induced fat malabsorption in human volunteers. *Clin Transl Gastroenterol* 2016;7:161.
60. Huang CB, George B, Ebersole JL. Antimicrobial activity of n-6, n-7 and n-9 fatty acids and their esters for oral microorganisms. *Arch Oral Biol* 2010;55:555–560.
61. Cândido FG, Valente FX, Grzeszkowiak LM, Moreira APB, Rocha DMUP, Alfenas RCG. Impact of dietary fat on gut microbiota and low-grade systemic inflammation: Mechanisms and clinical implications on obesity. *Int J Food Sci Nutr* 2018;69:125–143.
62. Yao CK, Muir JG, Gibson PR. Review article: insights into colonic protein fermentation, its modulation and potential health implications. *Aliment Pharmacol Ther* 2016;43:181–196.
63. Blachier F, Beaumont M, Portune KJ, Steuer N, Lan A, Audebert M, et al. High-protein diets for weight management: interactions with the intestinal microbiota and consequences for gut health. A position paper by the my new gut study group. *Clin Nutr* 2019; 38:1012–1022.
64. Centre for Reviews and Dissemination Systematic Reviews CRD's guidance for undertaking reviews in health care. York: Centre for Reviews and Dissemination, University of York; 2009; Available at: [https://www.york.ac.uk/media/crd/Systematic\\_Reviews.pdf](https://www.york.ac.uk/media/crd/Systematic_Reviews.pdf).
65. Coughlan M, Cronin P. Doing a literature review in nursing, health and social care. 2nd edn. London: SAGE 2013.
66. Gough D, Oliver S, Thomas J. An introduction to systematic reviews. 2nd edn. London: SAGE 2017.
67. Moher D, Liberati A, Tetzlaff J, Altman DG. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *PLoS Med* 2009;6:1000097.
68. National Institute of Care and Excellence (NICE) Methods for the development of NICE public health guidance. 2018 Available at: <https://www.nice.org.uk/process/pmg4/chapter/introduction>.
69. Aveyard H. Doing a literature review in health and social care: a practical guide. 4th edn. Maidenhead: Open University Press 2019.
70. Braun V, Clarke V. Successful qualitative research: A practical guide for beginners. 1st edn. London: SAGE 2013.
71. Higgins JPT, Green S. (eds.) *Cochrane Handbook for Systematic Reviews of Interventions Version 5.1.0*. 2011. Available at: <https://training.cochrane.org/handbook>.
72. Brinkworth GD, Noakes M, Clifton PM, Bird AR. Comparative effects of very low-carbohydrate, high-fat and high-carbohydrate, low-fat weight-loss diets on bowel habit and faecal short-chain fatty acids and bacterial populations. *Br J Nutr* 2009;101:1493–1502.
73. Duncan SH, Lohley GE, Holtrop G, Ince J, Johnstone AM, Louis P, et al. Human colonic microbiota associated with diet, obesity and weight loss. *Int J Obes* 2008;32:1720–1724.
74. Mardinoglu A, Wu H, Bjornson E, Zhang C, Hakkarainen A, Räsänen SM, et al. An integrated understanding of the rapid metabolic benefits of a carbohydrate-restricted diet on hepatic steatosis in humans. *Cell Metab* 2018;27:559–571.
75. Murtaza N, Burke LM, Vlahovich N, Charlesson B, O' Neill H, Ross ML, et al. The effects of dietary pattern during intensified training on stool microbiota of elite race walkers. *Nutrients* 2019;11:261.
76. Russell WR, Gratz SW, Duncan SH, Holtrop G, Ince J, Scobbie L, et al. High-protein, reduced-carbohydrate weight-loss diets promote metabolite profiles likely to be detrimental to colonic health. *Am J Clin Nutr* 2011;93:1062–1072.
77. Swidsinski A, Dörffel Y, Loening-Baucke V, Gille C, Göktas Ö, Reißhauer A, et al. Reduced mass and diversity of the colonic microbiome in patients with multiple sclerosis and their improvement with ketogenic diet. *Front Microbiol* 2017;8:1141.
78. Gopalakrishnan S, Ganeshkumar P. Systematic reviews and meta-analysis: Understanding the best evidence in primary healthcare. *J Family Med Prim Care* 2013;2:9-14.
79. Duncan SH, Belenguer A, Holtrop G, Johnstone AM, Flint HJ, Lohley GE. Reduced dietary intake of carbohydrates by obese subjects results in decreased concentrations of butyrate and butyrate-producing bacteria in feces. *Appl Environ Microbiol* 2007;73:1073–1078.
80. Newell C, Bomhof MR, Reimer RA, Hittel DS, Rho JM, Shearer J. Ketogenic diet modifies the gut microbiota in a murine model of autism spectrum disorder. *Molecular Autism* 2016;7:37.

81. Zhang Y, Zhou S, Zhou Y, Yu L, Zhang L, Wang Y. Altered gut microbiome composition in children with refractory epilepsy after ketogenic diet. *Epilepsy Research* 2018;145:163–168.
82. Knudsen KEB, Nørskov NP, Bolvig AK, Hedemann MS, Laerke HN. Dietary fibres and associated phytochemicals in cereals. *Mol Nutr Food Res* 2017;61:1600518.
83. 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;7:979.
84. de Vries W, Stouthamer AH. Pathway of glucose fermentation in relation to the taxonomy of Bifidobacteria. *J Bacteriol* 1967;93:574–576.
85. De Vuyst L, Moens F, Selak M, Rivière A, Leroy F. Summer Meeting 2013: growth and physiology of Bifidobacteria. *J Appl Microbiol* 2014;116:477–491.
86. Puchalska P, Crawford PA. Multi-dimensional Roles of Ketone Bodies in Fuel Metabolism, Signaling, and Therapeutics. *Cell Metab* 2017;25:262–284.
87. Tognini P, Murakami M, Liu Y, Eckel-Mahan KL, Newman JC, Verdin E, et al. Distinct circadian signatures in liver and gut clocks revealed by ketogenic diet. *Cell Metab* 2017;26:523–538.
88. Ang QY, Alexander M, Newman JC, Tian Y, Cai J, Upadhyay V, et al. Ketogenic diets alter the gut microbiome resulting in decreased intestinal Th17 cells. *Cell* 2020;181:1263–1275.
89. Baxter NT, Schmidt AW, Venkataraman A, Kim KS, Waldron C, Schmidt TM. Dynamics of human gut microbiota and short-chain fatty acids in response to dietary interventions with three fermentable fibres. *mBio* 2019;10:02566–18.
90. Simpson HL, Campbell BJ. Review article: dietary fibre-microbiota interactions. *Aliment Pharmacol Ther* 2015;42:158–179.
91. Duncan SH, Louis P, Thomson JM, Flint HJ. The role of pH in determining the species composition of the human colonic microbiota. *Environ Microbiol* 2009;11:2112–2122.
92. Holscher HD. Dietary fibre and prebiotics and the gastrointestinal microbiota. *Gut Microbes* 2017;8:172–184.
93. Russell DA, Ross RP, Fitzgerald GF, Stanton C. Metabolic activities and probiotic potential of Bifidobacteria. *Int J Food Microbiol* 2011;149:88–105.
94. van der Beek CM, Dejong CHC, Troost FJ, Masclee AAM, Lenaerts K. Role of short-chain fatty acids in colonic inflammation, carcinogenesis, and mucosal protection and healing. *Nutr Rev* 2017;75:286–305.
95. Peng L, Li ZR, Green RS, Holzman IR, Lin J. Butyrate enhances the intestinal barrier by facilitating tight junction assembly via activation of AMP-activated protein kinase in Caco-2 cell monolayers. *J Nutr* 2009;139:1619–1625.
96. Jung TH, Park JH, Jeon WM, Han KS. Butyrate modulates bacterial adherence on LS174T human colorectal cells by stimulating mucin secretion and MAPK signaling pathway. *Nutr Res Pract* 2015;9:343–349.
97. Corrêa-Oliveira R, Fachi JL, Vieira A, Sato FT, Vinolo MAR. Regulation of immune cell function by short-chain fatty acids. *Clin Transl Immunol* 2016;5:73.
98. Ríos-Covián D, Ruas-Madiedo P, Margolles A, Gueimonde M, De los Reyes-Gavilán CG, Salazar N. Intestinal short chain fatty acids and their link with diet and human health. *Front Microbiol* 2016;7:185.
99. Huda-Faujan N, Abdulmir AS, Fatimah AB, Anas OM, Shuhaimi M, Yazid AM, et al. The impact of the level of the intestinal short chain fatty acids in inflammatory bowel disease patients versus healthy subjects. *Open Biochem J* 2010;4:53–58.
100. Kumari R, Ahuja V, Paul J. Fluctuations in butyrate-producing bacteria in ulcerative colitis patients of North India. *World J Gastroenterol* 2013;19:3404–3414.
101. Owczarek D, Rodacki T, Domagała-Rodacka R, Cibor D, Mach T. Diet and nutritional factors in inflammatory bowel diseases. *World J Gastroenterol* 2016;22:895–905.
102. Rapozo DCM, Bernardazzi C, De Souza HSP. Diet and microbiota in inflammatory bowel disease: the gut in disharmony. *World J Gastroenterol* 2017;23:2124–2140.
103. Yang J, Yu J. The association of diet, gut microbiota and colorectal cancer: what we eat may imply what we get. *Protein Cell* 2018;9:474–487.
104. Matsuoka K, Kanai T. The gut microbiota and inflammatory bowel disease. *Semin Immunopathol* 2015;37:47–55.
105. Wang T, Cai G, Qiu Y, Fei N, Zhang M, Pang X, et al. Structural segregation of gut microbiota between colorectal cancer patients and healthy volunteers. *ISME J* 2012;6:320–329.
106. Bottacini F, Ventura M, van Sinderen D, Motherway MOC. Diversity, ecology and intestinal function of bifidobacteria. *Microbial Cell Factories* 2014;13:4.
107. Ling X, Linglong P, Weixia D, Hong W. Protective effects of *Bifidobacterium* on intestinal barrier function in LPS-induced enterocyte barrier injury of Caco-2 monolayers and in a rat NEC model. *PLoS ONE* 2016;11:0161635.
108. O’Callaghan A, van Sinderen D. Bifidobacteria and their role as members of the human gut microbiota. *Front Microbiol* 2016;7:925.
109. Coakley M, Ross RP, Nordgren M, Fitzgerald G, Devery R, Stanton C. Conjugated linoleic acid biosynthesis by human-derived *Bifidobacterium* species. *J Appl Microbiol* 2003;94:138–145.
110. Le Leu RK, Hu Y, Brown IL, Woodman RJ, Young GP. Synbiotic intervention of *Bifidobacterium lactis* and resistant starch protects against colorectal cancer development in rats. *Carcinogenesis* 2010;31:246–251.
111. Sivan A, Corrales L, Hubert N, Williams JB, Aquino-Michaels K, Earley ZM, et al. Commensal *Bifidobacterium* promotes antitumor immunity and facilitates anti-PD-L1 efficacy. *Science* 2015;350:1084–1089.
112. Borges-Canha M, Portela-Cidade JP, Dinis-Ribeiro M, Leite-Moreira AF, Pimentel-Nunes P. Role of colonic microbiota in colorectal carcinogenesis: a systematic review. *Rev Esp Enferm Dig* 2015;107:659–671.
113. Gianfredi V, Salvatori T, Villarini M, Moretti M, Nucci D, Realdon S. Is dietary fibre truly protective against colon cancer? a systematic review and meta-analysis. *Int J Food Sci Nutr* 2018;69:904–915.

114. Yao Y, Suo T, Andersson R, Cao Y, Wang C, Lu J, et al. Dietary fibre for the prevention of recurrent colorectal adenomas and carcinomas. *Cochrane Database Syst Rev* 2017;1:003430.
115. Sze MA, Topçuoğlu BD, Lesniak NA, Ruffin MT, Schloss PD. Faecal short-chain fatty acids are not predictive of colonic tumor status and cannot be predicted based on bacterial community structure. *mBio* 2019;10:01454-19.
116. Hou JK, Abraham B, El-Serag H. Dietary intake and risk of developing inflammatory bowel disease: a systematic review of the literature. *Am J Gastroenterol* 2011;106:563–573.
117. Wong C, Harris PJ, Ferguson LR. Potential benefits of dietary fibre intervention in inflammatory bowel disease. *Int J Mol Sci* 2016;17:919.
118. Rahman M, Muhammad S, Khan MA, Chen H, Ridder DA, Müller-Fielitz H, et al. The  $\beta$ -hydroxybutyrate receptor HCA2 activates a neuroprotective subset of macrophages. *Nat Commun* 2014;5:3944.
119. Stilling RM, van de Wouw M, Clarke G, Stanton C, Dinan TG, Cryan JF. The neuropharmacology of butyrate: The bread and butter of the microbiota-gut-brain axis?. *Neurochem Int* 2016;99:110–132.
120. Shimazu T, Hirschey MD, Newman J, He W, Shirakawa K, Le Moan N, et al. Suppression of oxidative stress and  $\beta$ -OHB as endogenous histone deacetylase. *Science* 2013;339:211–214.
121. Newman JC, Verdin E.  $\beta$ -hydroxybutyrate: Much more than a metabolite. *Diabetes Res Clin Pract* 2014;106:173–181.
122. Klement RJ, Paziienza V. Impact of different types of diet on gut microbiota profiles and cancer prevention and treatment. *Medicina* 2019;55:84.
123. Arbolea S, Watkins C, Stanton C, Ross RP. Gut *bifidobacteria* populations in human health and aging. *Front Microbiol* 2016;7:1204.
124. Furet JP, Kong LC, Tap J, Poitou C, Basdevant A, Bouillot JL, et al. Differential adaptation of human gut microbiota to bariatric surgery-induced weight loss: links with metabolic and low-grade inflammation markers. *Diabetes* 2010;59:3049–3057.
125. Wu X, Ma C, Han L, Nawaz M, Gao F, Zhang X, et al. Molecular characterisation of the faecal microbiota in patients with type II diabetes. *Curr Microbiol* 2010;61:69–78.
126. Million M, Angelakis E, Maraninchi M, Henry M, Giorgi R, Valero R, et al. Correlation between body mass index and gut concentrations of *Lactobacillus reuteri*, *Bifidobacterium animalis*, *Methanobrevibacter smithii* and *Escherichia coli*. *Int J Obes* 2013;37:1460–1466.
127. Gao X, Jia R, Xie L, Kuang L, Feng L, Wan C. Obesity in school-aged children and its correlation with Gut *E.coli* and *Bifidobacteria*: a case-control study. *BMC Pediatr* 2015;15:64.
128. Aizawa E, Tsuji H, Asahara T, Takahashi T, Teraishi T, Yoshida S, et al. Possible association of *Bifidobacterium* and *Lactobacillus* in the gut microbiota of patients with major depressive disorder. *J Affect Disord* 2016;202:254–257.
129. Tseng CH, Wu CY. The gut microbiome in obesity. *J Formos Med Assoc* 2019;118:3–9.
130. Xiong Y, Miyamoto N, Shibata K, Valasek MA, Motoike T, Kedzierski RM, et al. Short-chain fatty acids stimulate leptin production in adipocytes through the G protein-coupled receptor GPR41. *Proc Natl Acad Sci U S A* 2004;101:1045–1050.
131. Cohen MM. Role of leptin in regulating appetite, neuroendocrine function, and bone remodeling. *Am J Med Genet* 2006;140:515–524.
132. Samuel BS, Shaito A, Motoike T, Rey FE, Backhed F, Manchester JK, et al. Effects of the gut microbiota on host adiposity are modulated by the short-chain fatty-acid binding G protein-coupled receptor, Gpr41. *Proc Natl Acad Sci U S A* 2008;105:167616772.
133. Tolhurst G, Heffron H, Lam YS, Parker HE, Habib AM, Diakogiannaki E. Short-chain fatty acids stimulate glucagon-like peptide-1 secretion via the G-protein-coupled receptor FFAR2. *Diabetes* 2012;61:364–371.
134. D'Alessio D. Intestinal hormones and regulation of satiety: the case for CCK, GLP-1, PYY, and Apo A-IV. *J Parent Enter Nutr* 2008;32:567–568.
135. Den Besten G, Van Eunen K, Groen AK, Venema K, Reijngoud DJ, Bakker BM. The role of short-chain fatty acids in the interplay between diet, gut microbiota, and host energy metabolism. *J Lip Res* 2013;54:2325–2340.
136. Cani PD, Amar J, Iglesias MA, Poggi M, Knauf C, Bastelica D, et al. Metabolic endotoxemia initiates obesity and insulin resistance. *Diabetes* 2007;56:1761–1772.
137. Cani PD, Bibiloni R, Knauf C, Neyrinck AM, Delzenne NM. Changes in gut microbiota control metabolic diet-induced obesity and diabetes in mice. *Diabetes* 2008;57:1470–1481.
138. Lyte JM, Gabler NK, Hollis JH. Postprandial serum endotoxin in healthy humans is modulated by dietary fat in a randomized, controlled, cross-over study. *Lipids Health Dis* 2016;15:186.
139. Mehta NN, McGillicuddy FC, Anderson PD, Hinkle CC, Shah R, Pruscino L, et al. Experimental endotoxemia induces adipose inflammation and insulin resistance in humans. *Diabetes* 2010;59:172–181.
140. Amar J, Burcelin R, Ruidavets JB, Cani PD, Fauvel J, Alessi MC, et al. Energy intake is associated with endotoxemia in apparently healthy men. *Am J Clin Nutr* 2008;87:1219–1223.
141. Maes M, Kubera M, Leunis JC, Berk M. Increased IgA and IgM responses against gut commensals in chronic depression: further evidence for increased bacterial translocation or leaky gut. *J Affect Disord* 2012;141:55–62.
142. Fuke N, Nagata N, Suganuma H, Ota T. Regulation of gut microbiota and metabolic endotoxemia with dietary factors. *Nutrients* 2019;11:2277.
143. Wang J, Tang H, Zhang C, Zhao Y, Derrien M, Rocher E, et al. Modulation of gut microbiota during probiotic-mediated attenuation of metabolic syndrome in high fat diet-fed mice. *ISME J* 2015;9:1–15.
144. Mazloom K, Siddiqi I, Covasa M. Probiotics: How effective are they in the fight against obesity?. *Nutrients* 2019;11:1–24.
145. Tan TG, Sefik E, Geva-Zatorsky N, Kua L, Naskar D, Teng F, et al. Identifying species of symbiont bacteria from the human gut that, alone, can induce intestinal Th17 cells in mice. *Proc Natl Acad Sci U S A* 2016;113:8141–8150.

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146. Flux MC, Lowry CA. Finding intestinal fortitude: Integrating the microbiome into a holistic view of depression mechanisms, treatment, and resilience. *Neurobiol Dis* 2020;135:104578.
147. Scriven M, Dinan T, Cryan J, Wall M, et al. Neuropsychiatric disorders: Influence of gut microbe to brain signalling. *Diseases* 2018;6:78.
148. Bonaz B, Bazin T, Pellissier S. The vagus nerve at the interface of the microbiota-gut-brain axis. *Front Neurosci* 2018;12:49.
149. Duman RS, Sanacora G, Krystal JH. Altered connectivity in depression: GABA and glutamate neurotransmitter deficits and reversal by novel treatments. *Neuron* 2019;102:75–90.
150. Barrett E, Ross RP, O'Toole PW, Fitzgerald GF, Stanton C.  $\gamma$ -Aminobutyric acid production by culturable bacteria from the human intestine. *J Appl Microbiol* 2012;113:411–417.
151. Boonstra E, de Kleijn R, Colzato LS, Alkemade A, Forstmann BU, Nieuwenhuis S. Neurotransmitters as food supplements: The effects of GABA on brain and behavior. *Front Psychol* 2015;6:1520.
152. Johnson KVA, Foster KR. Why does the microbiome affect behaviour?. *Nat Rev Microbiol* 2018;16:647–655.
153. Bercik P, Park AJ, Sinclair D, Khoshdel A, Lu J, Huang X, et al. The anxiolytic effect of *Bifidobacterium longum* NCC3001 involves vagal pathways for gut-brain communication. *J Neurogastroenterol Motil* 2011;23:1132–1139.
154. Desbonnet L, Garrett L, Clarke G, Bienenstock J, Dinan TG. The probiotic *Bifidobacteria infantis*: an assessment of potential antidepressant properties in the rat. *J Psychiatr Res* 2008;43:164–174.
155. Pinto-Sanchez MI, Hall GB, Ghajar K, Nardelli A, Bolino C, Lau JT, et al. Probiotic *Bifidobacterium longum* NCC3001 reduces depression scores and alters brain activity: a pilot study in patients with Irritable Bowel Syndrome. *Gastroenterol* 2017;153:448–459.
156. la Rosa PS, Brooks JP, Deych E, Boone EL, Edwards DJ, Wang Q, et al. Hypothesis testing and power calculations for taxonomic-based human microbiome data. *PLoS ONE* 2012;7:52078.
157. Kelly BJ, Gross R, Bittinger K, Sherrill-Mix S, Lewis JD, Collman R, et al. Power and sample-size estimation for microbiome studies using pairwise distances and PERMANOVA. *Bioinformatics* 2015;31:2461–2468.
158. Debelius J, Song SJ, Vazquez-Baeza Y, Xu ZZ, Gonzalez A, Knight R. Tiny microbes, enormous impacts: What matters in gut microbiome studies?. *Genome Biol* 2016;17:217.
159. Staley C, Kaiser T, Khoruts A. Clinician guide to microbiome testing. *Dig Dis Sci Springer US*, 2018;63:3167–3177.
160. Monda V, Villano I, Messina A, Valenzano A, Esposito T, Moscatelli F, et al. Exercise modifies the gut microbiota with positive health effects. *Oxid Med and Cell Longev* 2017:3831972.
161. Nettleton JE, Reimer RA, Shearer J. Reshaping the gut microbiota: impact of low calorie sweeteners and the link to insulin resistance?. *Physiol Behav* 2016;164:488–493.
162. McFarland LV. Use of probiotics to correct dysbiosis of normal microbiota following disease or disruptive events: a systematic review. *BMJ Open* 2014;4:005047.
163. Derrien M, van Hylckama Vlieg JE. Fate, activity, and impact of ingested bacteria within the human gut microbiota. *Trends Microbiol* 2015;23:354–366.