

Review

# New Insights in Prebiotic Utilization: A Systematic Review

Martina Arapović<sup>1</sup>, Leona Puljić<sup>1</sup>, Nikolina Kajić<sup>1</sup> , Brankica Kartalović<sup>2</sup> , Kristina Habschied<sup>3</sup>   
and Krešimir Mastanjević<sup>3,\*</sup> 

<sup>1</sup> Faculty of Agriculture and Food Technology, University of Mostar, Biskupa Čule bb, 88000 Mostar, Bosnia and Herzegovina; martina.arapovic@aptf.sum.ba (M.A.); leona.puljic@aptf.sum.ba (L.P.); nikolina.kajic@aptf.sum.ba (N.K.)

<sup>2</sup> Institut Biosens, Zorana Đinđića 1, 21000 Novi Sad, Serbia; brankica.kartalovic@biosense.rs

<sup>3</sup> Faculty of Food Technology Osijek, J.J. Strossmayera University of Osijek, F. Kuhača 18, 31000 Osijek, Croatia; khabschi@ptfos.hr

\* Correspondence: kmastanj@gmail.com

**Abstract:** The hectic pace of modern life often leads to quick solutions, both in lifestyle and the choice of food we consume. The importance of the gut microbiome and its balance is being increasingly researched, with the prebiotic concept itself becoming a topic of scientific investigation. The aim of this paper is to analyze scientific studies on the understanding of prebiotics conducted between 2019 and 2024 in order to see what new knowledge, new sources, new ways of use, and newly established effects on certain disease states have been discovered during this period. The question that the authors are trying to answer is how specific prebiotics affect the growth and activity of selected probiotic strains in the human gut (have impact on gut microbiome) and what the implications of these interactions are. Four databases were searched: Pubmed/MEDLINE, Springerlink, Google Scholar, and Scopus. The keywords used were prebiotics, functional food, probiotics, gut microbiome, and trends. A systematic review of 30 scientific studies on the topic of prebiotics revealed significant advances in understanding and application. Research particularly indicates how prebiotics stimulate the growth of beneficial probiotic strains, such as *Lactiseibacillus rhamnosus*, *Lactiplantibacillus plantarum*, and *Bifidobacterium*. In addition, innovative approaches in food production, including pasta rich in prebiotic fibers, chocolate with inulin and stevia, and the use of fruit by-products, show promising results in creating “healthier” food options. Although the papers had differing objectives and research methodologies, certain similarities were found. All papers emphasized the importance of using prebiotics, although it depended on the type they come from and their impact on the gut microbiome, i.e., the stimulation of probiotic action within the gut microbiome, which consequently has benefits on health. This review serves as a springboard for further research in this exciting field, with the ultimate goal of harnessing the power of prebiotics to improve health outcomes.

**Keywords:** prebiotic; probiotics; gut microbiome; functional food



**Citation:** Arapović, M.; Puljić, L.; Kajić, N.; Kartalović, B.; Habschied, K.; Mastanjević, K. New Insights in Prebiotic Utilization: A Systematic Review. *Processes* **2024**, *12*, 867.

<https://doi.org/10.3390/pr12050867>

Academic Editor: Dariusz Dziki

Received: 5 April 2024

Revised: 18 April 2024

Accepted: 23 April 2024

Published: 25 April 2024



**Copyright:** © 2024 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

## 1. Introduction

“Let food be thy medicine, and medicine be thy food” (Hippocrates)

In the last decade, prebiotics have attracted significant attention in scientific research due to their role in stimulating the intestinal microflora in the degradation of nutrients and improving the overall health status of humans. Their degradation products are short-chain fatty acids (SCFA) that are released into the bloodstream, consequently affecting not only the gastrointestinal tract but also other distant organs [1]. Through the effort and work of scientists, prebiotics today, after numerous studies thanks to the Consensus Panel of the International Scientific Association for Probiotics and Prebiotics (ISAPP), are defined as follows: “Substrates that are selectively utilized by host microorganisms conferring a health benefit” [2]. The concept of prebiotics was first introduced by Glenn Gibson and Marcel Roberfroid in 1995 [3]. Only a few compounds from the group of carbohydrates,

such as short-chain and long-chain  $\beta$ -fructans (FOS and inulin) and lactulose or galactooligosaccharides (GOS), can be classified as prebiotics. By modulating the gut microbiota, prebiotics stimulate the growth of “good bacteria” probiotics, such as *Lactobacillus* and *Bifidobacterium*, while reducing the growth of those “bad bacteria” or pathogens [4]. By emphasizing research and raising people’s awareness of proper and balanced nutrition, the problem of the onset of related diseases is sought to be suppressed. Thus, the aim of this paper is to analyze the latest findings on the prebiotic potential and answer the question of how specific prebiotics affect the growth and activity of selected strains of probiotics in digestion (in vitro and in vivo) and what the implications of these interactions are. This will be investigated by reviewing the literature and studying papers in databases relevant to the given subject of research. Also, the aim is to identify key knowledge gaps in the field of prebiotics and topics that are currently of interest to scientists, such as optimal dosages, their interactions with probiotics that potentially result in the greatest health benefits, and similar topics.

## 2. Existing Knowledge: Types of Prebiotics, Their Sources, and Impact

In the next section, we will cover the most frequently mentioned types of prebiotics and their sources that have been mentioned in further research. In addition to the mentioned ones, other known groups of potential prebiotics are oligosaccharides derived from starch and glucose, where resistant starch belongs, which is an indigestible component of human intestines, and other oligosaccharides, such as pectin (pectin oligosaccharide—POS), for example, flavanols from cocoa, which belong to oligosaccharides without carbohydrates and have shown their potential by acting on the stimulation of lactic acid bacteria.

### 2.1. Fructans

Fructans are a type of prebiotic composed of inulin and fructooligosaccharides (FOS) or oligofructose. Their structure is a linear chain of fructose with a  $\beta$  (2→1) bond. They usually consist of glucose units with a  $\beta$  (2→1) bond at the end [5]. Initial research pointed to the selective stimulation of fructans on lactic acid bacteria, but in recent years, more and more emphasis is being placed on chain length as an important factor influencing the determination of the type of bacteria that can ferment [6].

#### 2.1.1. Inulin

Inulin-type fructans are polymers made from fructose linked to terminal  $\alpha$ -linked glucose via a  $\beta$  (2→1) bond. Chicory is considered the richest source of inulin. Several studies have shown that inulin-type fructans can enhance the growth of bifidobacteria and *Anaerostipes* [7]. Inulin is most commonly used as a fat substitute [8]; it has been proven to enhance mineral absorption in the body [9,10], alleviate constipation [11], and promote the growth and development of *Lactobacillus* and *Bifidobacterium* strains within the colon [12], which is why it is important to this study.

#### 2.1.2. FOS

Fructooligosaccharides (FOS) are a researched type of prebiotic that can be found in bananas, wheat, onions, and garlic. The main component of the FOS structure in  $\beta$  (2→1) bonds is fructose. FOS derivatives are beneficial for colon health, as they promote the selective growth of bifidobacteria and lactobacilli and potentially inhibit microorganisms in intestinal cells, enhance the maintenance of body bacterial balance, alleviate constipation, and maintain healthy gut flora [13].

### 2.1.3. GOS

Galactooligosaccharides (GOSs), a product of lactose degradation, are classified into two subgroups: GOS with an excess of galactose at C3, C4, or C6 and GOS produced from lactose by enzymatic transglycosylation. The end product of this reaction is mainly a mixture of tri- to pentasaccharides with galactose in  $\beta$  (1 $\rightarrow$ 6),  $\beta$  (1 $\rightarrow$ 3), and  $\beta$  (1 $\rightarrow$ 4) bonds. This type of GOS is also called trans-galacto-oligosaccharides or TOS [14]. GOS is considered one of the most researched types of prebiotics and is safe for consumption. Galactooligosaccharides can greatly stimulate the growth of bifidobacteria and lactobacilli [15]. Its most common use currently is in infant milk formula, where it has proven to be a help in gut modulation by promoting the growth of *Bifidobacterium* strains in infants who cannot be fed breast milk [16].

### 2.2. Newly Recognized Unclassified Prebiotics

With the advancement of technology, prebiotic preparation methods have been optimized. In addition, various new types of prebiotics have been developed (mainly including polysaccharides, polyphenols, and polypeptide polymers) [17]. Emerging prebiotics are mainly found in algae, fruit juices, and fruit such as chokeberry [18]. Although knowledge about them is not yet as established as about prebiotic types FOS and GOS, their potential deserves in-depth study and has a promising future. The mentioned research is located in Section 4.

### 2.3. Importance of Prebiotic Interaction on Probiotic Growth and Stimulation Mechanism

The human digestive system, often described as the “second brain,” plays a crucial role in maintaining health. The gut microbiome consists of approximately  $3.9 \times 10^{13}$  bacteria in the large intestine, composed of probiotics, neutral bacteria, and pathogenic bacteria [19], which compete with each other to establish a balance in order to prevent the development of diseases. Scientists are constantly searching for new sources of prebiotics and probiotics and their synergistic action, as diseases related to gut balance disorders, such as obesity, type 2 diabetes, irritable bowel syndrome, various mental disorders, and colon cancer, are in constant worrying increase. The notion of probiotic food often brings to mind mild kefir and fermented dairy food [20]. The gut microbiome is in constant interaction with the human body, helping the body digest and absorb nutrients from food, metabolize toxic waste produced in the intestines, and produce functional substances necessary for life. This primarily refers to amino acids, vitamins, short-chain fatty acids (SCFA), and other substances [21]. Since probiotics are live microorganisms that enhance gut function with sufficient intake, their growth and reproduction require the intake of mainly polysaccharides that the body cannot digest—prebiotics. After the intake of a prebiotic compound, it passes through the small intestine and reaches the large intestine undegraded. Here, probiotic microorganisms break them down primarily into SCFAs, which later act on the organism, using fermentation. *Firmicutes* and *Bacteroidetes* are physiologically dominant species in the fecal microbiota of the adult large intestine, while *Actinobacteria* predominates in infants [22]. The ratio of *Firmicutes* to *Bacteroides* was often considered a trigger for obesity, but newer meta-analyses refute this theory. Unlike studies done on mice, human studies have not confirmed these results. Disagreements between these studies highlight the lack of reliable microbial taxonomic indicators for obesity [23]. Further research is needed to establish this fact. Furthermore, research indicates that the composition of the gut microbiome may be related to personality traits [24]. Among probiotic strains, the most commonly described are strains of *Lactobacillus* (belongs to the phylum *Firmicutes*) and *Bifidobacterium* (belongs to the phylum *Actinobacteria*) [22]. In addition to them, this group also includes *Enterococcus*, *Streptococcus*, and *Escherichia*. The fungus *Saccharomyces boulardii* is not naturally found in the gut microbiome, but its supplementation is of particular interest to people who need to replace gut dysbiosis caused by the use of antibiotics [25].

### 3. Materials and Methods

Due to PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses), guidelines were followed in this review. Search was performed from inception until January 2024 in Pubmed/Medline, Springerlink, Google Scholar, and Scopus databases for randomized clinical trials that assessed the effects of prebiotic type on human gut in vitro and in vivo among adults and children and that were published in English language.

#### 3.1. Inclusion Criteria

The research encompasses insights into the effects of prebiotics based on clinical trials, specifically randomized controlled trials (RCTs). The target population was adults and children with specific health conditions, comparing the composition of the gut microbiota and its impact on health before and after the study. The studies include comparisons of the type of prebiotic sources used and their impact on health. The consequences of prebiotic action, including improvement, stimulation of probiotic growth, and the duration of the study, are taken into account. The time span of the studies considered was within the last 5 years (2019–2024).

#### 3.2. Exclusion Criteria

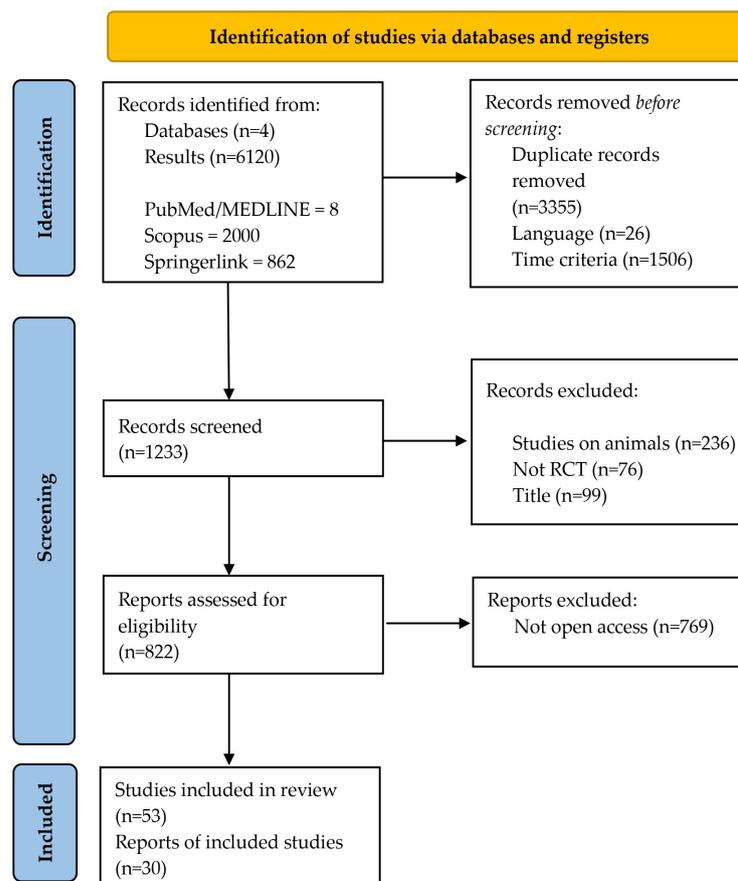
All studies not conducted on humans were excluded (animals, review papers, study protocols, or observational studies). Case studies were also not included. Clinical trials that did not have a precisely defined type of prebiotic used and an examination of their subsequent action either on the gut microbiome or on the stimulation of probiotic strain growth in the gut microbiome were not considered. Papers that did not belong to this study by title, i.e., included a type of research that is not the subject of inclusion, were excluded. The duration of the studies was not limited.

#### 3.3. Research Process

Four databases were utilized for the search: PubMed/MEDLINE, Scopus, Springerling, and Google Scholar. The keywords used for Google Scholar were “prebiotics”, “functional food”, “probiotics”, “gut microbiome”, “trends”. During the Scopus database search, the “Food Science” filter was applied. After the search, a database of 6120 scientific papers was obtained. Prior to the search, papers that overlapped ( $n = 3355$ ), were not originally written in English ( $n = 26$ ), and did not belong to the range of research subjects by year ( $n = 1506$ ) were excluded. Through the processing of the obtained papers ( $n = 1233$ ), studies conducted on animals ( $n = 236$ ), non-randomized controlled trials ( $n = 76$ ), those not relevant to the title of this systematic review ( $n = 99$ ), and those without access enabled ( $n = 592$ ) were excluded. Inclusive, this study covered 53 literature sources, of which 30 scientific papers were taken for analysis.

### 4. Results and Discussion

The investigated articles are listed in Tables 1 and 2, and the PRISMA flowchart is presented in Figure 1. Based on the stated inclusion criteria for the research, the authors have arrived at the following observations.



**Figure 1.** PRISMA flow diagram used for research.

#### 4.1. Impact of Prebiotics from New Sources on the Growth of Specific Probiotics

The act of choosing and utilizing food does not need to be intricate. In their research, [26] tested the ways how different concentrations of apple, banana, and mango peel powder affect the growth of probiotics. In this way, they tried to utilize the by-products of fruit processing. They concluded that a small amount of purified fruit peel powder at a concentration of 2% shows prebiotic action by stimulating the growth of *Lactocaseibacillus rhamnosus* (LGG), *L. casei*, and *B. animalis subslactis* (BB-12), proving that the by-product can be commercially utilized. Furthermore, researching purified banana peel powder, [27] enriched cookies with fibers while controlling the growth rate parameter of the probiotic strain *Lactobacillus* spp. They found a significantly higher number of colonies compared to the controlled groups of inulin, glucose, and the placebo. Further research is anticipated on the organoleptic assessment of this type of cookie. Aiming to exploit the potential of radish leaf by-products, ref. [28] investigated its potentials as prebiotics. Specifically, plant extracts show promise as potential therapeutics for the prevention of metabolic syndrome and obesity. The composition of immature radish extract, i.e., the prebiotic activity of RGP (radish green polysaccharide), was analyzed. Prebiotic action was established, resulting in changes in the concentration of SCFA and the pH of the gut microbiome. Probiotic strains successfully utilize RGP, proving its potential as a prebiotic. In addition, research conducted with oral consumption of the probiotic strain *Lactiplantibacillus plantarum* P-8, in combination with RGP, resulted in an increase in the probiotic strain and the inhibition of fat accumulation in adipocytes. The final results led to a reduction in fat accumulation in the body, which could effectively reduce obesity. This study proves that RGP, as a polysaccharide, promises as a natural prebiotic ingredient in the food industry. With the aim of proving polyphenols as prebiotics, in a 12-week study conducted on a group of healthy young men [18], the subjects consumed aronia extract or whole aronia fruit powder,

which had been previously purified. The research findings indicate a positive impact on the potential consumption of polyphenols from aronia for maintaining the cardiovascular health of the young men studied. In an attempt to demonstrate the prebiotic effect of five types of legume honey and the probiotic effect on *L. rhamnosus* and *L. paracasei*, ref. [29] used astragalus, carob, alfalfa, sainfoin, and indigo honey in their research. They concluded that the polyphenols present in these types of honey show a prebiotic potential on *L. rhamnosus* and *L. paracasei* strains with antagonistic action towards the five most dangerous pathogens depending on the type of honey. The authors look forward to further research on the postbiotic action of legume honey. Fucoïdan (purity  $\geq 95\%$ ), isolated from brown algae, attracted the attention of [30], so they investigated its prebiotic potential. In their research, finding an optimal concentration of 8 mg/mL, they proved the regulation of the antibacterial ability of *L. rhamnosus* and noticed a great concentration dependence on the action. They observed the growth inhibition zone. Unlike them, ref. [31] concluded that the water extract of *P. ostreatus*, obtained via extraction from white oyster mushroom, stands out due to its  $\beta$ -glucan content. Namely, water-soluble  $\beta$ -glucan from *P. ostreatus* proved a prebiotic potential by stimulating the growth of *L. plantarum*, creating a symbiotic. Ref. [32] investigated the prebiotic properties of a sample of purified rice bran extract (RB) on the stimulation of the growth and the inhibition of gastrointestinal probiotic pathogens. As a parallel control, they used inulin and distilled water. They followed the growth of the genera *L. casei* and *L. plantarum*. The results of this study indicate that the rice bran extract obtained by xylanase (RB1) contains a higher concentration of oligosaccharides that serve as a prebiotic compound alongside those obtained with water (RB2). This study predicts another possibility of using by-products from the food industry with the aim of creating functional food. Trying to mitigate the negative effect of sugar in chocolate but not to eliminate it from the diet, ref. [33] conducted research on chocolate with a low content of fat, sugar, phytochemicals, and prebiotics, making chocolate from cocoa, inulin, and stevia. The more acceptable version of chocolate proved to give the best sensory experience to consumers based on the hedonic scale. Furthermore, the polyphenols, flavonoids, and alkaloids coming from cocoa powder were noted as good sources of phytochemicals and contributed to the benefits. The addition of inulin together with phytonutrients gave the product prebiotic properties and stimulated the growth of *L. rhamnosus*. In addition, inulin was used to limit the caloric value acting as a good substitute for fat. The glycemic index was limited since stevia was used as a sweetener. This product promises to be a healthier, prebiotically richer version of chocolate, and future research is expected.

**Table 1.** Impact of prebiotics from new sources on the growth of specific probiotics.

Author	Prebiotic Source	Concentrations	Probiotic Type	Impact
Hafza Fasiha Zahid et al. (2021) [26]	Apple, banana, and mango peel powder	0%, 2%, and 4%	<i>Lactocaseibacillus rhamnosus</i> , <i>L.casei</i> , <i>Bifidobacterium lactis</i>	↑ lactic acid bacteria (LAB).
Chee Yee Tan et al. (2024) [27]	Banana peel powder, Plantain Peel Powder, inulin	10%, 20%, and 30%	<i>Lactobacillus</i> spp.	↑ <i>Lactobacillus</i> spp.
Florinda Fratianni et al. (2023) [29]	Legumes honey's	-	<i>Lactocaseibacillus casei</i> , <i>Lactobacillus gasseri</i> , <i>Lactocaseibacillus paracasei</i> subsp. <i>Paracasei</i> , <i>Lactiplantibacillus plantarum</i> , <i>Lactocaseibacillus rhamnosus</i>	Improve prebiotic properties; have inhibitory biofilm effect

Table 1. Cont.

Author	Prebiotic Source	Concentrations	Probiotic Type	Impact
Yanli Zhu et al. (2021) [30]	Fucoidan as prebiotic, isolated from <i>Undaria pinnatifida</i>	0, 0.8, 8, 80 mg/mL,	<i>L. rhamnosus</i>	Inhibiting bacterial infections
Anindita Deb Pal et al. (2023) [33]	Prebiotic Enriched (inulin) Chocolates	2% chocolate extract and 1% sub-cultured broth	<i>Lacticaseibacillus rhamnosus</i>	May promote well-being consequent to alternative of conventional chocolates
Yu Ra Lee et al. (2023) [28]	Radish ( <i>Raphanus sativus</i> L.)	4.9%	<i>B. bifidum</i> , <i>B. Longum</i> , <i>Lacticaseibacillus paracasei</i> , <i>Lactiplantibacillus plantarum</i> , <i>Escherichia coli</i> , <i>Lactobacillus lactis</i>	↑ <i>Lacticaseibacillus</i> , <i>Lactobacillus</i> , and <i>Bifidobacterium</i> ↑ SCFA
Ryan Haryo Setyawan et al. (2023) [31]	White Oyster Mushroom ( <i>P. ostreatus</i> ) Extract	2% w/v and 0.25% w/v	<i>Lactiplantibacillus plantarum</i> and pathogen <i>Escherichia coli</i>	↑ <i>L. Plantarum</i> Dad-13
Istas et al. (2019) [18]	Aronia berry whole fruit (WF), extract (EX),	500 mg/capsule/day	↑ in <i>Anaerostipes</i> ↑ in <i>Bacteroides</i>	Significantly improved endothelial function
Thornthan Sawangwani et al. (2024) [32]	Rice bran extract	1% v/v	<i>L. casei</i> , <i>L. Plantarum</i> , and pathogen <i>B. cereus</i> , <i>E. coli</i>	↑ <i>L. casei</i> , <i>L. plantarum</i> ;; high potential for inhibiting the growth of pathogenic <i>B. cereus</i> and <i>E. coli</i> .

↑—increasing value.

#### 4.2. Correlation of the Use of Prebiotics from New Sources and Human Health Status

##### 4.2.1. The Impact of Consuming Fructans as Prebiotics

In an attempt to prevent the diseased states of the “modern age”, there is increasing research in the correlation between gut microbiota, its composition, and its impact on human health. Hectic lifestyles make it difficult to properly choose and access “healthy” foods, so researchers are trying to discover new, modify existing, and make “healthier” versions of popular foods. In the search for healthy food, ref. [34] researched the impact of dragon fruit oligosaccharides on gut microbiota. The research proved prebiotic activity and the modulation of the gut microbiome. A small dose of just 4 g/day had an impact on IgA, while a dose of 8 g/day successfully promotes the growth of the probiotic strains *Bifidobacterium* spp. and *Faecalibacterium* while inhibiting the growth of *Escherichia coli* strains. Conducting a double-blind RCT study, where FOS was used as a placebo and a parallel encapsulated combination of FOS with *Lactiplantibacillus plantarum* and *Bifidobacterium breve*, ref. [35] interestingly, concluded that despite increased fat intake, markers associated with obesity improved in the group where the combined version of prebiotics and probiotics was consumed. This evidence provides important information on the possible use of prebiotics in the prevention of obesity, but further research is still required. Investigating the correlation between the intakes of the antipsychotic olanzapine in individuals suffering from schizophrenia and weight gain, ref. [36] attempted to prevent this side effect. By setting up two studies, they examined the effect of olanzapine on the consumption of the probiotic itself and olanzapine and probiotic with a combination of 10 g of bitter melon (*Momordica charantia*) and oligosaccharides. It was concluded that this combination effectively reduces the weight gain caused by the use of olanzapine while retaining the desired psychopathological effects. Probiotics alone were not sufficient to prevent the side effect of weight gain caused by the use of this drug. Also, it was discovered that the addition

of a combination of probiotics and prebiotic fibers of bitter melons have a significant benefit for insulin resistance. Striving to make a modified and “healthier” version of pasta, ref. [37] proved to be able to lower the glycemic index by adding barley  $\beta$ -glucans and probiotic spores BC30. This study shows for the first time that basic food such as pasta can be modified into a symbiotic food that can have beneficial effects in people at risk of developing diseases due to an unhealthy lifestyle. This opens up possibilities for designing new “healthier” alternatives to basic foods. Unlike [37], in the research of [38], the impact of barley  $\beta$ -glucans on lipid metabolism in patients at high risk of developing metabolic syndrome was monitored. B-glucans influenced the formation of short-chain fatty acids, a change in the composition of the gut microbiota with an increase in the diversity of microorganisms in accordance with previous observations conducted on animal models, primarily *Bifidobacteriae*. The need for a “personalized” approach to nutrition has also been concluded. Comparing the prebiotic properties of  $\beta$ -glucans from oats and caffeine-free extracts from green coffee on obesity and its comorbidities, ref. [39], conducting research on individuals with excessive body weight, concluded that regular consumption of the four nutraceuticals based on the combination of oat beta glucans and prebiotics from green coffee may reduce cardiovascular risk and help in preventing type 2 diabetes and obesity. By supplementing oligofructose-enriched p-inulin, ref. [40] researched its impact on the metabolic response of the gut in people with chronic kidney disease (CKD). The results suggest a reduction in carbohydrate metabolism in the subjects, which led to a change in the gut microbiome and consequently resulted in an increase in *Bifidobacteria* and *Anaerostipes*. Trying to create a non-pharmacological drug for people suffering from respiratory diseases, ref. [41] researched the impact of adding inulin to inflammatory indicators in obese people with asthma. They proved the potential of using inulin alone, unlike that with the addition of probiotics, which interestingly had fewer benefits than inulin alone. The authors look forward to further research on this non-pharmacological drug. Pointing to the persistence of the link between strength and muscle function in older people and the use of prebiotics, ref. [42] gave their subjects a prebiotic supplement (inulin and fructo-oligosaccharide). Healthy older twin pairs were used in the examination. They concluded the prebiotic effect on improving cognition in the elderly, but they did not have an impact on strength and muscle function compared to the placebo, as expected. With their research on the impacts of adding the prebiotic fiber inulin (fructooligosaccharide), Vitafiber (isomaltooligosaccharide), and Fibremax (mixture of different fiber) to the diet of healthy women, [43] proved that the greatest impact was from the addition of inulin on the intestinal microflora and the increase in the strain *Bifidobacterium*. Further research is needed to individually determine the selection of the most suitable prebiotic fibers for maximum utilization in the formation of short-chain fatty acids.

#### 4.2.2. The Impact of Consuming GOS as Prebiotics

An extremely high-quality study proved the prebiotic potential of their objects of research, GOS, MOS, GMOS, and GMPS, by [44] showing prebiotic effects for *C. butyricum* and bifidobacteria. In conclusion, different prebiotic effects of galactosyl and mannosyl carbohydrates have been proven, as well as the production of SCFA and potential symbiotic effects, and a connection to their physical–chemical characteristics has been established. An in vivo study, compared to commercial GOS and MOS, showed that hydrolyzed galactomannans with specific degrees of polymerization have special advantages in prebiotic effects and display potential as new prebiotic products. The first study that followed the correlation between the intake of prebiotic galactooligosaccharides (GOS) and the gut microbiome of children with autism was conducted by [45]. A significant increase in the probiotic strain *Bifidobacterium* was determined after consumption when compared to the placebo group, but there were no changes in the spectrum of children’s behavior. Although there was no significant difference in the results between the groups, the absence of side effects and the discovered medium effects of the study were encouraging and require further research. Ref. [46], in their double-blind, placebo-controlled, 4-week study of galac-

tooligosaccharide (GOS) supplement interventions, sought to characterize the impact of GOS on indices of emotional well-being and gut microbial composition in a sample of participants at the end of adolescence and in early adulthood. It was found that GOS increases the abundance of *Bifidobacterium* in 4 weeks in tandem with a reduced manifestation of anxiety and indications of modifying attention bias in a sample of adolescent women. The presented data promise that GOS could be effective in influencing the reduction of anxiety.

#### 4.2.3. Newly Recognized Unclassified Prebiotics and Their Impact on Health

In an attempt to exploit pectooligosaccharides (POS) from lemon peels as prebiotics, ref. [47], compared with the standard prebiotic fructooligosaccharides (FOS) in older people by researching in vitro fermentation, proved a greater potential of POS in the production of short-chain fatty acids (SCFA) and a smaller proportion of the formation of branched-chain fatty acids (BCFA) from the FOS group. The potential prebiotic properties of black and white pepper were investigated by [48] compared with inulin as a positive control, and it was concluded that black and white pepper shows significantly higher prebiotic properties by promoting the growth of bifidobacteria compared to inulin. The authors attribute this to the component of arabinogalactan present in *Piper nigrum*. In line with the research on prebiotic potentials of spices, ref. [49] concluded that the substrates present in *Curcuma longa* (turmeric), *Zingiber officinale* (ginger), *Piper longum* (pipli or long pepper), and *Piper nigrum* (black pepper) may drive beneficial alterations in gut communities, thereby altering their collective metabolism and contributing to the salubrious effects on digestive efficiency and health. Researching the potential of black rice [50] as a prebiotic, the authors concluded that the black rice extract contains 71.9 mg of anthocyanins per 100 g of dry weight of the extract, so it could be used as a prebiotic ingredient in the gut microbiota in obese people. The research was prompted by the fact that the consumption of 70.7 mg of anthocyanins daily during 8 weeks of consumption favorably affects the composition of gut bacteria. It has been established that there is a connection between high levels of anthocyanins to a reduction in inflammatory markers and changes in gut microbiota and the prevention of obesity in a way that produces strains of *Lachnospiraceae* and *Ruminococaceae*. These strains in turn produce butyrate, which plays a crucial role in increasing the protection of the intestinal barrier, thereby inhibiting the entry of anti-inflammatory molecules into the bloodstream and preventing the onset of metabolic endotoxemia. By researching the potential of arabinoxylan-oligosaccharides (AXOS) from wheat bran extract, ref. [51] found that the intake of wheat bran extract increases fecal *Bifidobacterium* and softens the consistency of the stool without major effects on energy metabolism in healthy people with slow GI transit. By proving the potential of resistant starch as a prebiotic, ref. [52] discovered the benefits of resistant corn starch that are associated with changes in the composition of the gut microbiota. These changes in the gut microbiota affect the bile acid profile, reduce inflammation by restoring the intestinal barrier, and inhibit lipid absorption. As a potential prebiotic polyphenol from *Montgomery cherry*, ref. [53] demonstrated an increase in *Lactobacillus* strains in 22/28 participants, but the examined middle-aged human population did not have significant changes in insulin responses as expected. However, there is still a lack of information in the literature about optimal bioactive doses for modifying the gut microbiome and managing the glucose response.

**Table 2.** Correlation of the use of prebiotics from new sources and human health status.

Author	A Source of Prebiotics	Participants	Type of Research	Dose	Period	Conclusions
Nattha Pansai et al. (2023) [34]	Dragon fruit oligosaccharides (DFO)	107	Randomized Double-Blind, Placebo Controlled Study	4 g and 8 g	4 weeks	↑ <i>Bifidobacterium</i> spp.; decreased harmful bacteria, especially, <i>Escherichia coli</i> . DFO improved the immune system (serum Ig A) at even a low dose (4 g/day)
Eun-Ji Song et al. (2020) [35]	Fructo-oligosaccharide as prebiotic; <i>B. breve</i> CBT BR3, <i>L. plantarum</i> CBT LP3 as probiotic	50	Double-blind, placebo-controlled, randomized clinical trial	2 tablets	12 weeks	Significantly improved obesity-related markers in obese people
Michael B. Sohn et al. (2024) [40]	Oligofructose-enriched inulin (p-inulin)	15	Nonrandomized, open-label, 3-phase pilot trial, with repeated measures within each phase	8 g/twice a day	28 weeks	↑ <i>Bifidobacterium</i> and <i>Anaerostipes</i> ; increase in abundance of microbial metabolites derived from carbohydrate metabolism.
Huang et al. (2022) [36]	Probiotic: Bifico containing live <i>Bifidobacterium</i> , <i>Lactobacillus</i> and <i>Enterococcus</i> Prebiotic: bitter melon ( <i>Momordica charantia</i> ) and oligosaccharides (fructooligosaccharides and oligoisomaltoses), kudzu starch, insulin and resistant dextrin	Study 1, 90 Study 2, 60	Two sequential, randomized clinical trials	Study 1: olanzapine (15–20 mg/day) plus probiotics (840 mg twice daily) vs. olanzapine monotherapy Study 2: olanzapine (15–20 mg/day) plus probiotics (840 mg twice daily) and dietary fibre (30 g twice daily) vs. olanzapine monotherapy	12 weeks	Probiotics plus dietary fibre significantly attenuated olanzapine-induced weight gain; Psychopathological symptoms improved in all groups

Table 2. Cont.

Author	A Source of Prebiotics	Participants	Type of Research	Dose	Period	Conclusions
Beatriz Míguez et al. (2020) [47]	Pectooligosaccharides obtained from lemon peel (POS)	6	In vitro experiments	6.5 g/day of FOS, POS or SIEM	3 days	↑ SCFA and the lowest BCFA; results support the potential of pectin-derived oligosaccharides as prebiotic candidates targeting gut health in the elderly
Mattea Müller et al. (2020) [51]	Wheat bran extract Arabinoxylan Oligosaccharide (AXOS)	48	Randomized, Placebo-Controlled, Double-blind study	15 g/day AXOS	12 weeks	↑ <i>Bifidobacterium</i> and softens stool
Mary Ni Lochlainn et al. (2024) [42]	Prebiotic supplement (inulin and fructo-oligosaccharides)	36 twin pairs (72 individuals)	Placebo Controlled Double-Blinded Randomised Controlled Trial	7.5 g of prebiotic: inulin (3.375 mg) and FOS (3.488 mg)	12 weeks	May improve cognition in ageing population
Jian Tan et al. (2023) [43]	Inulin (fructo-oligosaccharide), Vitafiber (isomalto-oligosaccharide), and Fibremax (mixture of different fiber)	28	Cross-over intervention Study	Three times daily, a total of 15 g of Vitafiber, 34 g of Fibrema, or 15 g of inulin	3 weeks	Inulin supplementation had highest impact on SCFA production.
Rebecca McLoughlin et al. (2019) [41]	Soluble fibre supplementation (inulin) with a probiotic	17	Randomised, double-blinded, placebo controlled 3-way cross-over trial	3 × day (inulin 12 g/day), soluble fibre + probiotic (inulin 12 g/day+ multi-strain probiotic N25 billion CFU) and placebo.	1 week	Improvements in airway inflammation, asthma control and gut microbiome composition following inulin supplementation
Donato Angelino et al. (2019) [37]	Whole-grain pasta containing barley β-glucans and <i>Bacillus coagulans</i> BC30, 6086	41	Single-Blind, Parallel, Randomized, Placebo-controlled Dietary Intervention Study	1 serving/d	12 weeks	Affected glycemia- and lipid-related markers; resistin in a subgroup of healthy obese or hyperglycemic volunteers

Table 2. Cont.

Author	A Source of Prebiotics	Participants	Type of Research	Dose	Period	Conclusions
Ana Velikonja et al. (2019) [38]	Barley beta glucans	43	Double-blind, placebo-controlled, randomised clinical trial	6 g/day	4 weeks	Affect lipid metabolism in patients with high risk for MS development; effect on glucose metabolism was not confirmed
Raquel Mateos et al. (2022) [39]	Oat Beta-Glucan; hydroxycinnamates, caffeine-free extract from green coffee	60	Randomized, dose-response, parallel, blind study	3 g d <sup>-1</sup> or 5 g d <sup>-1</sup> doses of 35% or 70% BG and a fixed amount of GCBE providing 600 mg d <sup>-1</sup> of phenols.	6 weeks	May reduce the cardiovascular risk and help in preventing type 2 diabetes and obesity
Siti Maisarah Nashri et al. (2023) [48]	<i>Piper nigrum</i> L.	3	In vitro colon fermentation compared with inulin effect	-	-	↑ <i>Bifidobacterium</i> spp., and <i>Lactobacillus/Enterococcus</i> , showed the ability to suppress colonic pathogen strain <i>Cl. histolyticum</i>
Christine T. Peterson et al. (2019) [49]	Curcuma longa (turmeric), Zingiber officinale (ginger), Piper longum (pipili or long pepper), and Piper nigrum (blackpepper)	12	In vitro anaerobic fecal cultivation	1%	-	May drive beneficial alterations in gut communities thereby altering their collective metabolism to contribute to the salubrious effects on digestive efficiency and health
Lear et al. (2019) [53]	Montmorency cherry (MC) drink	28	RCT, DB, placebo	30 mL 2×/day AC—296 mg/day PP—1040 mg/day	4 weeks	Significant ↓ in insulin No significance—blood glucos. ↑ <i>Lactobacillus</i> spp.
Rachmat Faisal Syamsu et al. (2023) [50]	Black Rice Extract ( <i>Oryza Sativa</i> L.)	33	Randomized control trial, placebo research with a pre-post test approach	Black Rice Extract 5.6 g/day	4 weeks	High anthocyanins so that it can be used as a prebiotic ingredient for the intestinal microbiota in obese subjects

Table 2. Cont.

Author	A Source of Prebiotics	Participants	Type of Research	Dose	Period	Conclusions
Huating Li et al. (2024) [52]	RS derived from maize (HAM-RS2, Hi-Maize 260 resistant starch)	37	Randomized placebo-controlled crossover design trial	40 g/day RS	8 weeks	RS-induced changes in the gut microbiota alter the bile acid profile, reduce inflammation by restoring the intestinal barrier; inhibit lipid absorption
Jacqueline K. Palmer et al. (2024) [45]	GOS (GOSYAN® GOS) supplementation	33	A double-blind randomised, placebo controlled trial	2.4 g/d	6 weeks	↑ <i>Bifidobacterium</i> but in this instance, resulted in only marginal effects on GI symptoms
Xinyi Wei et al. (2020) [44]	Hydrolyzed guar gum (GMOS), manno-oligosaccharide (MOS), and galacto-oligosaccharide (GOS)	3	In vitro fermentation	-	-	Individualized prebiotic effects which are associated with their chemical structures including their glycoside composition
Nicola Johnstone et al. (2021) [46]	Galacto-oligosaccharides (Biotis™ GOS, ≈7.5 g powder~5.5 g GOS)	64	Double-blind placebo-controlled trial	7.5 g/day	4 weeks	↑ <i>Bifidobacterium</i> ; data presented are indicative that GOS may be effective in influencing the expression of anxiety

↑—increasing value. ↓—decreasing value.

### 4.3. The Limitations of the Reviewed Studies

Disadvantages that can be observed in this review, which the authors also highlighted, were primarily the heterogeneity of prebiotics. Namely, prebiotics are diverse and isolated from different sources, and their chemical structure and effect can vary. This complicates the comparison and generalization of results from different studies. Furthermore, methodological problems are highlighted, namely that research on prebiotics is often conducted on small samples with different doses and durations, which, given the lack of standardized protocols, can complicate the comparison of results. There is also a lack of long-term studies that are necessary for a better understanding of their effects on health. While there is evidence of short-term benefits, there is a lack of research tracking long-term outcomes, such as disease prevention. Finally, every person, and therefore the composition of their gut microbiota, is unique, so some people may benefit while others will not. It is important to note that prebiotics have the potential to improve overall health, however, future research should focus on addressing the aforementioned challenges in order to better understand the effects and optimal application.

## 5. Conclusions

A systematic review of 30 papers on the topic of prebiotics has revealed significant advancements in their understanding and application. The research particularly indicates that prebiotics promote the growth of beneficial probiotic strains such as *Lactobacillus rhamnosus*, *Lactiplantibacillus plantarum*, and *Bifidobacterium*. In addition, innovative approaches in food production, including pasta rich in prebiotic fibers, chocolate with inulin and stevia, and the utilization of fruit by-products, show promising results in creating “healthier” food options. These discoveries are especially relevant in the larger context of the global obesity epidemic, wherein prebiotics show a potential to reduce and prevent the inflammatory markers associated with this condition. Furthermore, new doors are opening for therapeutic interventions via discoveries of the connections between the gut microbiome and what has been often referred to as “our second brain”—the gut itself—as well the impact of this connection on regulating metabolism and mental health. This work not only confirms and highlights numerous benefits of prebiotics but also aims to stimulate the scientific community for further research to fully exploit their potential and health benefits. In the future, scientists should investigate the long-term effects of prebiotic supplementation and its impacts on various aspects of human health. The addition of new types of prebiotics to the classification is anticipated to orient scientists more towards new insights and types of prebiotic potential.

**Author Contributions:** Conceptualization, L.P. and M.A.; investigation, M.A. and N.K.; resources, K.M. and K.H.; writing—original draft preparation, M.A. and L.P.; writing—review and editing, K.M. and B.K.; supervision, K.M. and B.K. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research received no external funding.

**Conflicts of Interest:** The authors declare no conflicts of interest.

## References

1. Guo, C.; Huo, Y.J.; Li, Y.; Han, Y.; Zhou, D. Gut-brain axis: Focus on gut metabolites short-chain fatty acids. *World J. Clin. Cases* **2022**, *10*, 1754–1763. [[CrossRef](#)]
2. Gibson, G.R.; Hutkins, R.; Sanders, M.E.; Prescott, S.L.; Reimer, R.A.; Salminen, S.J.; Scott, K.; Stanton, C.; Swanson, K.S.; Cani, P.D.; et al. Expert consensus document: The International Scientific Association for Probiotics and Prebiotics (ISAPP) consensus statement on the definition and scope of prebiotics. *Nat. Rev. Gastroenterol. Hepatol.* **2017**, *14*, 491–502. [[CrossRef](#)]
3. Gibson, G.R.; Roberfroid, M.B. Dietary modulation of the human colonic microbiota: Introducing the concept of prebiotics. *J. Nutr.* **1995**, *125*, 1401–1412. [[CrossRef](#)] [[PubMed](#)]
4. Kailasapathy, K.; Chin, J. Survival and therapeutic potential of probiotic organisms with reference to *Lactobacillus acidophilus* and *Bifidobacterium* spp. *Immunol. Cell Biol.* **2000**, *78*, 80–88. [[CrossRef](#)]

5. Louis, P.; Flint, H.J.; Michel, C. How to Manipulate the Microbiota: Prebiotics. In *Microbiota of the Human Body, Advances in Experimental Medicine and Biology*; Schwierztz, A., Ed.; Springer: Cham, Switzerland, 2016; Volume 902. [[CrossRef](#)]
6. Scott, K.P.; Martin, J.C.; Duncan, S.H.; Flint, H.J. Prebiotic stimulation of human colonic butyrate-producing bacteria and bifidobacteria, in vitro. *FEMS Microbiol. Ecol.* **2014**, *87*, 30–40. [[CrossRef](#)]
7. Moens, F.; Verce, M.; De Vuyst, L. Lactate- and acetate-based cross-feeding interactions between selected strains of lactobacilli, Bifidobacteria and colon bacteria in the presence of inulin-type fructans. *Int. J. Food Microbiol.* **2017**, *241*, 225–236. [[CrossRef](#)]
8. Ahmed, W.; Rashid, S. Functional and therapeutic potential of inulin: A comprehensive review. *Crit. Rev. Food Sci. Nutr.* **2019**, *59*, 1–13. [[CrossRef](#)] [[PubMed](#)]
9. Wan, X.; Guo, H.; Liang, Y.; Zhou, C.; Liu, Z.; Li, K.; Niu, F.; Zhai, X.; Wang, L. The physiological functions and pharmaceutical applications of inulin: A review. *Carbohydr. Polym.* **2020**, *246*, 116589. [[CrossRef](#)] [[PubMed](#)]
10. Lin, H.; Wang, Q.; Yuan, M.; Liu, L.; Chen, Z.; Zhao, Y.; Das, R.; Duan, Y.; Xu, X.; Xue, Y.; et al. The prolonged disruption of a single-course amoxicillin on mice gut microbiota and resistome, and recovery by inulin, Bifidobacterium longum and fecal microbiota transplantation. *Environ. Pollut.* **2020**, *265*, 114651. [[CrossRef](#)]
11. Bastard, Q.L.; Chapelet, G.; Javaudin, F.; Lepelletier, D.; Montassier, E. The effects of inulin on gut microbial composition: A systematic review of evidence from human studies. *Eur. J. Clin. Microbiol. Infect. Dis.* **2020**, *39*, 403–413. [[CrossRef](#)]
12. Shoaib, M.; Shehzad, A.; Omar, M.; Rakha, A.; Raza, H.; Sharif, H.R. Inulin: Properties, health benefits and food applications. *Carbohydr. Polym.* **2016**, *147*, 444–454. [[CrossRef](#)] [[PubMed](#)]
13. Hussain, M.; Anjum, F.M.; Rahim, M.A.; Saeed, F.; Khalid, W. Functional and nutraceutical properties of fructo-oligosaccharides derivatives: A review. *Int. J. Food Properties* **2021**, *24*, 1588–1602. [[CrossRef](#)]
14. Macfarlane, G.; Steed, H.; Macfarlane, S. Bacterial metabolism and health-related effects of galacto-oligosaccharides and other prebiotics. *J. Appl. Microbiol.* **2008**, *104*, 305–344. [[CrossRef](#)] [[PubMed](#)]
15. Gibson, G.R.; Scott, K.P.; Rastall, R.A.; Tuohy, K.M.; Hotchkiss, A.; Dubert-Ferrandon, A.; Gareau, M.; Murphy, E.F.; Saulnier, D.; Loh, G. Dietary prebiotics: Current status and new definition. *Food Sci. Technol. Bull. Funct. Foods* **2010**, *7*, 1–19. [[CrossRef](#)]
16. Van Hoffen, E.; Ruiter, B.; Faber, J.; M'Rabet, L.; Knol, E.F.; Stahl, B. A specific mixture of short-chain galacto-oligosaccharides and long-chain fructooligosaccharides induces a beneficial immunoglobulin profile in infants at high risk for allergy. *Allergy* **2009**, *64*, 484–487. [[CrossRef](#)] [[PubMed](#)]
17. Rezende, E.S.V.; Lima, G.C.; Naves, M.M. Dietary fibers as beneficial microbiota modulators: A proposed classification by prebiotic categories. *Nutrition* **2021**, *89*, 111217. [[CrossRef](#)] [[PubMed](#)]
18. Ista, G.; Wood, E.; Le Sayec, M.; Rawlings, C.; Yoon, J.; Dandavate, V.; Cera, D.; Rampelli, S.; Costabile, A.; Fromentin, E.; et al. Effects of aronia berry (poly)phenols on vascular function and gut microbiota: A double-blind randomized controlled trial in adult men. *Am. J. Clin. Nutr.* **2019**, *110*, 316–329. [[CrossRef](#)] [[PubMed](#)]
19. Jones, J.L.; Foxx-Orenstein, A.E. The role of probiotics in inflammatory bowel disease. *Dig. Dis. Sci.* **2007**, *52*, 607–611. [[CrossRef](#)]
20. Arapović, M.; Puljić, L.; Kajić, N.; Banožić, M.; Kartalović, B.; Habschied, K.; Mastanjević, K. The Impact of Production Techniques on the Physicochemical Properties, Microbiological, and Consumer's Acceptance of Milk and Water Kefir Grain-Based Beverages. *Fermentation* **2024**, *10*, 2. [[CrossRef](#)]
21. You, S.; Ma, Y.; Yan, B.; Pei, W.; Wu, Q.; Ding, C.; Huang, C. The promotion mechanism of prebiotics for probiotics: A review. *Front. Nutr.* **2022**, *9*, 1000517. [[CrossRef](#)]
22. Mariarosaria, M. Bifidobacteria, Lactobacilli... when, how and why to use them. *Glob. Pediatr.* **2024**, *8*, 100139. [[CrossRef](#)]
23. Criton, V.J.; Joy, S. Beyond skincare routines: Follow your gut to healthy skin—A review of the interplay between gut microbiome and skin. *J. Ski. Sex. Transm. Dis.* **2023**, *12*, 1–7. [[CrossRef](#)]
24. Johnson, K.V. Gut microbiome composition and diversity are related to human personality traits. *Hum. Microbiome J.* **2020**, *15*, 100069. [[CrossRef](#)] [[PubMed](#)]
25. Mack, D.R. Probiotics-mixed messages. *Can. Fam. Physician* **2005**, *51*, 1455. [[PubMed](#)]
26. Zahid, H.F.; Ranadheera, C.S.; Fang, Z.; Ajlouni, S. Utilization of Mango, Apple and Banana Fruit Peels as Prebiotics and Functional Ingredients. *Agriculture* **2021**, *11*, 584. [[CrossRef](#)]
27. Tan, C.Y.; Arifin, N.N.M.; Sabran, R.M. Banana Peels as Potential Prebiotic and Functional Ingredient. *J. Gizi Pangan* **2024**, *19* (Suppl. 1), 119–126. [[CrossRef](#)]
28. Lee, Y.R.; Lee, H.-B.; Kim, Y.; Shin, K.-S.; Park, H.-Y. Prebiotic and Anti-Adipogenic Effects of Radish Green Polysaccharide. *Microorganisms* **2023**, *11*, 1862. [[CrossRef](#)]
29. Fratianni, F.; De Giulio, B.; d'Acerno, A.; Amato, G.; De Feo, V.; Coppola, R.; Nazzaro, F. In Vitro Prebiotic Effects and Antibacterial Activity of Five Leguminous Honeys. *Foods* **2023**, *12*, 3338. [[CrossRef](#)] [[PubMed](#)]
30. Zhu, Y.; Liu, L.; Sun, Z.; Ji, Y.; Wang, D.; Mei, L.; Shen, P.; Li, Z.; Tang, S.; Zhang, H.; et al. Fucooidan as a marine-origin prebiotic modulates the growth and antibacterial ability of Lactobacillus rhamnosus. *Int. J. Biol. Macromol.* **2021**, *180*, 599–607. [[CrossRef](#)]
31. Setyawan, R.H.; Saskiawan, I.; Widhyastuti, N.; Kasirah, K.; Mulyad, M. Prebiotic potency from White Oyster Mushroom (*Pleurotus ostreatus*) Extract. *Berrita Biol.* **2023**, *22*, 51–59. [[CrossRef](#)]
32. Sawangwan, T.; Kajadman, D.; Kulchananimit, R. Determination of prebiotic properties of rice bran extract. *Biosci. Microbiota Food Health* **2024**. [[CrossRef](#)]
33. Pal, A.D.; Chakraborty, P.; Gandhi, R.J. Nutritional and Functional Properties of Prebiotic Enriched Chocolates. *Indian J. Sci. Technol.* **2024**, *17*, 426–435. [[CrossRef](#)]

34. Pansai, N.; Detarun, P.; Chinnaworn, A.; Sangsupawanich, P.; Wichienchot, S. Effects of dragon fruit oligosaccharides on immunity, gut microbiome, and their metabolites in healthy adults—A randomized double-blind placebo controlled study. *Food Res. Int.* **2023**, *167*, 112657. [[CrossRef](#)] [[PubMed](#)]
35. Song, E.J.; Han, K.; Lim, T.J.; Lim, S.; Chung, M.; Nam, M.; Kim, H.; Nam, Y.D. Effect of probiotics on obesity-related markers per enterotype: A double-blind, placebo-controlled, randomized clinical trial. *EPMA J.* **2020**, *11*, 31–51. [[CrossRef](#)] [[PubMed](#)]
36. Huang, J.; Kang, D.; Zhang, F.; Yang, Y.; Liu, C.; Xiao, J.; Long, Y.; Lang, B.; Peng, X.; Wang, W.; et al. Probiotics Plus Dietary Fiber Supplements Attenuate Olanzapine-Induced Weight Gain in Drug-Naïve First-Episode Schizophrenia Patients: Two Randomized Clinical Trials. *Schizophr. Bull.* **2022**, *48*, 850–859. [[CrossRef](#)] [[PubMed](#)]
37. Angelino, D.; Martina, A.; Rosi, A.; Veronesi, L.; Antonini, M.; Mennella, I.; Vitaglione, P.; Grioni, S.; Brighenti, F.; Zavaroni, I.; et al. Glucose- and Lipid-Related Biomarkers Are Affected in Healthy Obese or Hyperglycemic Adults Consuming a Whole-Grain Pasta Enriched in Prebiotics and Probiotics: A 12-Week Randomized Controlled Trial. *J. Nutr.* **2019**, *149*, 1714–1723. [[CrossRef](#)] [[PubMed](#)]
38. Velikonja, A.; Lipoglavšek, L.; Zorec, M.; Orel, R.; Avguštin, G. Alterations in gut microbiota composition and metabolic parameters after dietary intervention with barley beta glucans in patients with high risk for metabolic syndrome development. *Anaerobe* **2019**, *55*, 67–77. [[CrossRef](#)] [[PubMed](#)]
39. Mateos, R.; García-Cordero, J.; Bravo-Clemente, L.; Sarriá, B. Evaluation of novel nutraceuticals based on the combination of oat beta-glucans and a green coffee phenolic extract to combat obesity and its comorbidities. A randomized, dose–response, parallel trial. *Food Funct.* **2022**, *13*, 574–586. [[CrossRef](#)]
40. Sohn, M.B.; Gao, B.; Kendrick, C.; Srivastava, A.; Isakova, T.; Gassman, J.J.; Fried, L.F.; Wolf, M.; Cheung, A.K.; Raphael, K.L.; et al. Pilot Studies in CKD Consortium. Targeting Gut Microbiome with Prebiotic in Patients With CKD: The TarGut-CKD Study. *Kidney Int. Rep.* **2023**, *9*, 671–685. [[CrossRef](#)]
41. McLoughlin, R.; Berthon, B.S.; Rogers, G.B.; Baines, K.J.; Leong, L.E.X.; Gibson, P.G.; Williams, E.J.; Wood, L.G. Soluble fibre supplementation with and without a probiotic in adults with asthma: A 7-day randomised, double blind, three way cross-over trial. *EBioMedicine* **2019**, *46*, 473–485. [[CrossRef](#)]
42. Ni Lochlainn, M.; Bowyer, R.C.E.; Moll, J.M.; García, M.P.; Wadge, S.; Baleanu, A.F.; Nessa, A.; Sheedy, A.; Akdag, G.; Hart, D.; et al. Effect of gut microbiome modulation on muscle function and cognition: The PROMOTE randomised controlled trial. *Nat. Commun.* **2024**, *15*, 1859. [[CrossRef](#)] [[PubMed](#)]
43. Tan, J.; Ribeiro, R.V.; Barker, C.; Daien, C.; De Abreu Silveira, E.; Holmes, A.; Macia, L. Functional profiling of gut microbial and immune responses toward different types of dietary fiber: A step toward personalized dietary interventions. *Gut Microbes* **2023**, *15*, 2274127. [[CrossRef](#)] [[PubMed](#)]
44. Wei, X.; Fu, X.; Xiao, M.; Liu, Z.; Zhang, L.; Mou, H. Dietary galactosyl and mannosyl carbohydrates: In-vitro assessment of prebiotic effects. *Food Chem.* **2020**, *329*, 127179. [[CrossRef](#)] [[PubMed](#)]
45. Palmer, J.K.; van der Pols, J.C.; Sullivan, K.A. A Double-Blind Randomised Controlled Trial of Prebiotic Supplementation in Children with Autism: Effects on Parental Quality of Life, Child Behaviour, Gastrointestinal Symptoms, and the Microbiome. *J. Autism Dev. Disord.* **2024**. [[CrossRef](#)] [[PubMed](#)]
46. Johnstone, N.; Milesi, C.; Burn, O. Anxiolytic effects of a galacto-oligosaccharides prebiotic in healthy females (18–25 years) with corresponding changes in gut bacterial composition. *Sci. Rep.* **2021**, *11*, 8302. [[CrossRef](#)] [[PubMed](#)]
47. Miguez, B.; Vila, C.; Venema, K.; Carlos Parajo, J.; Luis Alonso, J. Prebiotic effects of pectooligosaccharides obtained from lemon peel on the microbiota from elderly donors using an in vitro continuous colon model (TIM-2). *Food Funct.* **2020**, *11*, 9984–9999. [[CrossRef](#)] [[PubMed](#)]
48. Nashri, S.M.M.; Tan, H.Y.; Sarbini, S.R. Evaluation of Piper Nigrum L. As a Prebiotic Ingredient Using In Vitro Colon Model. *Borneo J. Resour. Sci. Technol.* **2023**, *13*, 13–23. [[CrossRef](#)]
49. Peterson, C.T.; Rodionov, D.A.; Iablokov, S.N.; Pung, M.A.; Chopra, D.; Mills, P.J.; Peterson, S.N. Prebiotic Potential of Culinary Spices Used to Support Digestion and Bioabsorption. *Evid. Based Complement. Altern. Med.* **2019**, *2019*, 8973704. [[CrossRef](#)]
50. Syamsu, R.F.; Bukhari, A.; Taslim, N.A.; Malik, S.G.; Wahyudin, E.; Bahar, B.; Daud, N.A. Husnaini Analysis of the Gut Microbiota in Obese Adolescent Subjects Fed Black Rice Extract (*Oryza Sativa* L.). *Azerbaijan Med. J.* **2023**, *63*, 04.
51. Müller, M.; Hermes, G.D.A.; Canfora Emanuel, E.; Holst, J.J.; Zoetendal, E.G.; Smidt, H.; Troost, F.; Schaap, F.G.; Damink, S.O.; Jocken, J.W.E.; et al. Effect of wheat bran derived prebiotic supplementation on gastrointestinal transit, gut microbiota, and metabolic health: A randomized controlled trial in healthy adults with a slow gut transit. *Gut Microbes* **2020**, *12*, 1704141. [[CrossRef](#)]
52. Li, H.; Zhang, L.; Li, J. Resistant starch intake facilitates weight loss in humans by reshaping the gut microbiota. *Nat. Metab* **2024**, *6*, 578–597. [[CrossRef](#)] [[PubMed](#)]
53. Lear, R.; O’Leary, M.; O’Brien Andersen, L.; Holt, C.C.; Stensvold, C.R.; van der Giezen, M.; Bowtell, J.L. Tart Cherry Concentrate Does Not Alter the Gut Microbiome, Glycaemic Control or Systemic Inflammation in a Middle-Aged Population. *Nutrients* **2019**, *11*, 1063. [[CrossRef](#)] [[PubMed](#)]

**Disclaimer/Publisher’s Note:** The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.