

Review



Alteration of the Intestinal Bacterial Community in Rainbow Trout (*Oncorhynchus mykiss*): The Role of Animal, Plant, and Microbial Diets

Aleksandra S. Chervochkina *, Nadezhda O. Pastukhova and Andrey S. Aksenov *D

Northern (Arctic) Federal University named after M.V. Lomonosov, Arctic Biomonitoring Laboratory, 163002 Arkhangelsk, Russia; n.pastuhova@narfu.ru

* Correspondence: a.chervochkina@narfu.ru (A.S.C.); a.s.aksenov@narfu.ru (A.S.A.)

Abstract: Intestinal bacterial community in rainbow trout (*Oncorhynchus mykiss*) is increasingly recognized as important for aquaculture and fish health. This review summarizes the current knowledge on how various feed components, including animal- and plant-derived ingredients, as well as other feed additives, influence the gut microbiota of rainbow trout. Studies using 16S rRNA gene profiling and metagenomics demonstrate how dietary changes affect the composition, diversity, and functionality of bacterial communities. The findings suggest that there is no single optimal alternative ingredient for fish feeds, but emphasize that the ratio of natural components plays a more significant role in shaping the intestinal bacterial community of trout. This balance is crucial for maintaining diversity, functionality, and overall fish health, thereby promoting growth and disease resistance, while enhancing aquaculture efficiency. The use of popular animal protein derived from the larva of *Hermetia illucens* increases the amount of Pseudomonadota, but the chitin they contain can prevent the absorption of nutrients. Thus, the development of optimal feed formulas requires careful selection and balancing of various natural ingredients to sustain a healthy microbiota and overall fish well-being.

Keywords: intestinal microbiota; rainbow trout; Oncorhynchus mykiss; dietary effects; 16S rRNA

1. Introduction

Rainbow trout (*Oncorhynchus mykiss*) is one of the most popular fish species for industrial farming. Compared to other commonly farmed species, such as Atlantic salmon, cod, and carp, rainbow trout stands out for its rapid growth rate and adaptability to various rearing conditions. It exhibits high productivity and capacity for rapid weight gain, making it competitive in the market. The nutritional value of rainbow trout meat is comparable to that of Atlantic salmon, being rich in protein and beneficial fatty acids. Trout is less demanding in terms of rearing conditions compared to cod and carp, making it a preferred choice for aquaculture [1,2]. Recent studies (2019–2024) have confirmed the popularity of trout for industrial farming and its value as a source of protein and fatty acids in the human diet.

The gut microbiome of fish significantly influences digestion, immune system development, and overall health. Studying the composition and functions of the intestinal bacterial communities in rainbow trout plays a key role in optimizing rearing conditions and enhancing production efficiency. Next-generation sequencing (NGS) technology allows for detailed analysis of the gut microbiome, identifying both dominant and rare, non-culturable microorganisms. Through NGS, in-depth studies of microbiome dynamics and their impact on digestion, the immune system, and the overall health of trout are made possible, contributing to the development of more effective rearing strategies. Modern techniques, such as DNA sequencing and metagenomics, have significantly enhanced the investigation of the gut microbiota in the rainbow trout. These methods enable the



Citation: Chervochkina, A.S.; Pastukhova, N.O.; Aksenov, A.S. Alteration of the Intestinal Bacterial Community in Rainbow Trout (*Oncorhynchus mykiss*): The Role of Animal, Plant, and Microbial Diets. *Microbiol. Res.* **2024**, *15*, 2199–2209. https://doi.org/10.3390/ microbiolres15040147

Academic Editor: Yiannis Kourkoutas

Received: 28 August 2024 Revised: 2 October 2024 Accepted: 25 October 2024 Published: 26 October 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/). identification of a broad spectrum of microorganisms, including non-culturable species, and allow for the analysis of their functions and interactions. These approaches provide more accurate assessments of the effects of a diet, probiotics, and rearing conditions on the microbiota, opening new avenues for optimizing aquaculture. Nutrition is a key factor in the life of the trout, as feed components significantly influence the composition of the gut microbiome, altering the balance of microorganisms and impacting the fish's vital functions. Therefore, studying the influence of feed components on the composition of bacterial communities in the fish intestine is of particular interest. Various animal and plant components in the diet can have diverse effects on its composition and function. Understanding these relationships will enable the development of optimal feed formulations that promote the health and growth of fish. The diversity of animal and plant components of rainbow trout feed will significantly influence the species composition of its gut microbiome, and certain feed components may foster a more favorable microbial community, which in turn could enhance fish health and production efficiency.

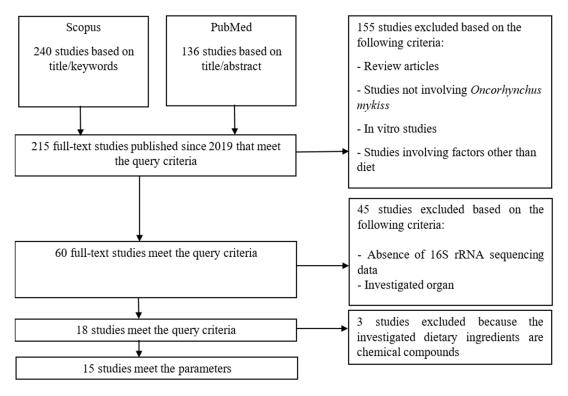
The NGS-based 16S rRNA sequencing method is more effective than traditional methods for studying fish intestinal microbiome, allowing for precise classification of bacteria, including rare species, and helping to track changes in the microbial community influenced by factors such as a diet [3]. The NGS-based 16S rRNA method has been widely applied in gut microbiota studies across various fish species, including carp, Atlantic salmon, and African catfish. These studies provide detailed insights into the microbiome, which is crucial for understanding its role in fish health and disease. For example, the study by Zhang et al. (2020) demonstrated that the microbiome of carp significantly changes depending on environmental conditions and diet, affecting its immune system and growth [4]. Similarly, the study by Wang et al. (2021) demonstrated that the microbiome of Atlantic salmon plays a key role in metabolism and nutrient absorption, which is crucial for aquaculture and productivity enhancement [5]. In turn, research on African catfish conducted by Lee et al. (2022) revealed that the microbiome of this species exhibits unique functional characteristics that could be utilized for the development of new disease prevention and treatment methods [6]. These studies underscore the importance of using NGS (Next-Generation Sequencing) for microbiome analysis, as this method allows for the identification of rare and difficult-to-culture microorganisms, as well as functional analysis of the microbiome.

Investigations into the microbiome of wild rainbow trout have shown that its gastrointestinal tract hosts diverse bacteria that play an essential role in digestion and the overall health of the fish. Bacillota (synonym of Firmicutes) and Bacteroidota (synonym of Bacteroidetes) are involved in the catabolism of complex carbohydrates and provide energy to the organism. Pseudomonadota (synonym of Proteobacteria) contribute to protein digestion and are involved in the immune response. Some pathogenic bacteria, such as *Aeromonas salmonicida* and *Flavobacterium psychrophilum*, can cause infections in fish, leading to diseases. Research indicates that these pathogenic bacteria can be found in the gastrointestinal tract of wild rainbow trout [7–11].

The aim of this review is to analyze and summarize current knowledge about the influence of various feed components, including animal, plant, and other natural additives, on the microbial composition of the gastrointestinal tract of rainbow trout, and to determine the most suitable raw materials for commercial feed production. To achieve this goal, an analysis of the diversity of the gastrointestinal microbiome of rainbow trout under different dietary regimes was conducted. This review will consolidate data on the relationship between nutrition and the microbiome, and provide practical recommendations for improving the conditions of rearing and breeding rainbow trout.

2. Methods

For the meta-analysis, studies were selected using the search terms gut OR intestine AND microbiome OR microbiota OR 'bacterial community' AND fish OR trout OR 'Oncorhynchus mykiss' in the abstracts, titles, and keywords within peer-reviewed databases



such as Scopus and PubMed. In Figure 1, a flowchart of the study selection process for conducting the analytical work is presented

Figure 1. Flow chart illustrating the systematic literature search and data selection processes.

A systematic selection process was conducted, starting with the establishment of stringent inclusion criteria followed by manual filtering of articles. The initial selection comprised 376 articles published within specific years, each providing full-text access. Each article was then meticulously reviewed based on its type, subject matter, and key research factors. Articles that did not involve dietary experiments with rainbow trout, as well as review papers lacking primary data, were excluded from the analysis.

Subsequent selection only included in vivo studies that involved analyses of various parts of the fish intestine, its contents, and mucus, with the infection status of the subjects not considered as an exclusion criterion.

To achieve the objectives of the study, only publications utilizing 16S rRNA bacterial profiling methods were included, narrowing the selection to 60 suitable articles. Of these, 42 articles were excluded as they did not meet the criteria for the specified methods. Further exclusions involved experiments focused on the effects of individual chemical compounds on the composition of the fish intestinal microbiota.

The selection of articles underwent rigorous filtration, with each exclusion justified on scientific grounds. Excluding articles that did not include dietary experiments with rainbow trout and meta-analyses was necessary to ensure a focus on primary data. The exclusion of studies that did not employ 16S rRNA bacterial profiling methods was driven by the need to obtain detailed and reliable data on the microbiota composition. Experiments involving individual chemical compounds were excluded as they did not provide a comprehensive analysis of the impact of diet.

The final review included 15 studies conducted under various fish rearing conditions, such as recirculating aquaculture systems, net pens, and flow-through aquarium systems. These studies described changes in the bacterial community composition of the gastrointestinal tract of rainbow trout in response to dietary modifications, whether involving complete replacement or partial substitution of specific ingredients.

3. Results

All the analyzed studies focused on identifying new raw materials to replace traditional ingredients, such as fishmeal, in commercial feeds with more environmentally sustainable and economically viable alternatives (Table 1). Experimental research was conducted across various countries, including Italy, China, France, Sweden, and New Zealand, both on farms and under laboratory conditions. The fish were reared in different systems, such as net pens, flow-through aquariums, and recirculating aquaculture systems (RAS). The microbial community in all studies was assessed by evaluating changes in the bacterial communities of the trout gastrointestinal tract using profiling of different hypervariable regions of the 16S rRNA gene.

Table 1. Studies with accessible data used for the analysis of the gut bacterial community composition of rainbow trout.

Feed Ingredient	Objects	Rearing System	Target Hypervariable Region	Results	Reference
Diet based on fishmeal and fish oil versus plant-derived	Intestinal	RAS	V5	The plant-based diet was dominated by Bacillota, while the traditional "marine diet" was dominated by Pseudomonadota, Bacteroidota, and Actinomycetota.	Hans-Christian Ingerslev et al., 2014 [12]
A diet with a different ratio of plant and animal components	Distal gut content	Tank	V3-V4	Dominance of <i>Cetobacterium</i> sp. in the microbiome of fish fed a diet high in plant protein and oils, compared to the marine diet.	Gatesoupe F-J et al., 2018 [13]
By-product meals from the rendering industry	Feces, proximal mucosa, distal mucosa	Flow- through	V3–V4	Bacillota, Pseudomanadota, Bacteroidota, Mycoplasma, and Actinomycetota taxa were predominant. The <i>Clostridia</i> taxon dominated in fish with a diet high in plant proteins. The addition of poultry by-products to the trout diet did not lead to changes in overall microbial diversity or richness. The observed changes in intestinal microbiome composition were actually related to the ratio of plant to animal proteins, regardless of the animal protein sources.	Rimoldi S et al., 2018 [14]
Black solider fly (Hermetia illucens)	Feces	RAS	V3–V4	The percentage of black soldier fly meal in the diet affected the abundance of Actinomycetota and Pseudomanadota (producers of lactic acid and butyrate), while maintaining an overall normal microbiome.	Terova, G. et al., 2019 [15]
Black solider fly (<i>Hermetia illucens</i>)	Gut content	Flow- through	V4	Replacing 50% of fish meal with black soldier fly meal in the diet reduced the numbers of Pseudomanadota, Lactobacillaceae, Leuconostocaceae, Fusobacteriaceae, and Shewanellaceae, and increased the numbers of Bacillota, <i>Vagococcus</i> , <i>Erysipelothrix</i> , and <i>Vibrio</i> . <i>Oceanobacillus</i> genus was present in the microbiome with black soldier fly meal diet.	Rimoldi, S. et al., 2021 [16]

Food Ingradiant	Objects	Rearing System	Target Hypervariable Region	Results	Reference
Feed Ingredient	Objects				
Mealworm (Tenebrio molitor)	Gut mucosa	Flow- through	V3–V4	Replacing fishmeal with <i>Tenebrio</i> <i>molitor</i> meal did not affect the composition and diversity of the microbiome. The only changes caused by the complete replacement of fishmeal with <i>Tenebrio molitor</i> larvae meal were a reduction in Pseudomonadota and a decrease in taxa belonging to the families Ruminococcaceae and Neisseriaceae.	Terova, G. et al., 2021 [17]
Neurospora intermedia	Gut content, mucosal scrapings	RAS	V4	Dominance of Tenericutes and Bacillota by the end of the experiment, compared to the traditional commercial diet.	Singh Aprajita et al., 2021 [18]
S. cerevisiae	Gut content, mucosa	Tank	V4	Yeast supplements did not disrupt the intestinal microbiota of trout.	D.L. et al., 2018 [19]
Capsaicin, Trametes versicolor, Ganoderma lucidum	Intestinal and gut content	Flow- through	V3–V4	The presence of capsaicin in the diet increased alpha diversity in the fish microbiome. Additionally, a diet containing only capsaicin increased the number of Clostridiaceae. With a <i>Ganoderma lucidum</i> diet, the abundance of Pseudomanadota increased.	Julia Mougin et al., 2023 [20]
Protein concentrate from cotton seeds	Gut content, mucosa	RAS	V3-V4	Replacing fish meal with cottonseed protein concentrate in the diet led to a decrease in Actinomycetota and an increase in Bacillota in the gut mucus microbiome.	Liu Yang et al., 2020 [21]
Plant-derived protein, rapeseed oil	Gut content	Cage	V3-V4	Replacing fish meal with plant-derived protein decreased Bacillota and Bacteroidetes, while Planctomycetota increased. Replacing fish meal and fish oil with plant alternatives increased the abundance of pathogens <i>Schlesneria</i> , <i>Brevundimonas</i> , and <i>Mycoplasma</i> .	Zhang, C. et al., 2023 [22]
A microcapsulated mixture of four organic acids and three essential oils	Proximal and distal intestines, feces	Tank	V3–V4	Overall, the experimental diet did not affect the alpha and beta diversity of the microbiome, but a therapeutic diet with organic plant acids and oils reduced the number of opportunistic pathogens <i>Aeromonas hydrophila</i> and <i>Acinetobacter</i> .	Huyben, D. et al., 2021 [23]
Chinese yam (<i>Dioscorea</i> <i>oppositifolia</i> L.) extract	Intestine and gut content	RAS	V3-V4	A diet containing 0.1% Chinese yam reduced Actinomycetota and <i>Patescibacteria</i> , while a diet containing 0.4% Chinese yam increased <i>Patescibacteria</i> , <i>Bifidobacteria</i> , <i>Marvinbryantia</i> , and <i>Candidatus</i> <i>Saccharimonas</i> .	F. et al., 2020 [24]

Table 1. Cont.

Feed Ingredient	Objects	Rearing System	Target Hypervariable Region	Results	Reference
Garlic extract	Intestinal mucosa	Cage	V4	In the control group, the most common genera were <i>Deefgea</i> and <i>Aeromonas</i> . In the experimental groups, the dominant genera were <i>Deefgea</i> and <i>Mycoplasma</i> . In the group receiving the highest amount of garlic extract, the dominant genus was <i>Aeromonas</i> , with <i>Deefgea</i> and <i>Exiguobacterium</i> also present.	Etyemez, Miray et al., 2028 [25]
A mix of plant proteins and oils, microalgae, Saccharomyces cerevisiae, Hermetia illucens	Gut content	Tank	V3-V4	A plant-based diet increased the abundance of Bacillota and decreased the abundance of Pseudomanadota. Replacing plant proteins with yeast and insect proteins restored the fish microbiome.	Pérez-Pascual, et al., 2021 [26]

Animal proteins in diets are being partially or entirely replaced with plant-based proteins, insect-derived proteins, and yeast fractions, while animal fats are substituted with essential oils and plant extracts. In some experiments, the diets included combination of alternative protein and fat sources alongside traditional formulations. In a study comparing marine and plant-based diets for rainbow trout's immune defense against *Yersinia ruckeri*, it was found that plant components exerted a prebiotic effect, stimulating the growth of specific bacteria. Notably, fish fed a plant-based diet under RAS conditions were dominated by Bacillota, including *Streptococcus, Leuconostoc*, and *Weissella*. In contrast, marine diets led to the dominance of Pseudomonadota, Bacteroidota, and Actinomycetota, demonstrating significant differences in the trout gut microbiota composition [12].

In another study, a group of scientists used food industry by-products combined with plant components as a replacement for fish meal in rainbow trout diets under aquarium conditions. The results indicated that changes in the microbial community of distal and proximal gut mucus, as well as feces, were associated not with the replacement of fish meal, but with the ratio of plant to animal components in the diet. When comparing marine diets to plant-based ones, it was observed that diets high in plant proteins and oils led to the dominance of *Cetobacterium* sp. in the microbiome, which synthesize vitamin B12 and support fish growth [13,14].

The primary alternative protein source is meals derived from the larvae of the insect *Hermetia illucens*. The inclusion of this meal in the diet significantly alters the microbial composition of the gastrointestinal community. Studies of gut content and feces have shown that diets containing varying amounts of black soldier fly meal under RAS conditions reduce the abundance of Actinomycetota and Pseudomonadota, which are producers of lactic acid and butyrate, and play a crucial role in the host's proper functioning. In flow-through systems, replacing 50% of fish meal with 15% black soldier fly meal also reduces the numbers of Pseudomonadota, Lactobacillaceae, Leuconostocaceae, Fusobacteriaceae, and Shewanellaceae, while increasing the abundance of Bacillota, *Vagococcus, Erysipelothrix*, and *Vibrio*. It was noted that the genus *Oceanobacillus* is observed in the microbiota only when *Hermetia illucens* is included in the diet. The use of meals derived from the larvae of another insect species, *Tenebrio molitor*, did not show significant changes in the gut mucus microbiome composition compared to commercial fishmeal-based feeds, according to weighted UniFrac analysis [15–17].

The use of yeast and fungi is not yet widespread, but several studies demonstrate the potential of these components as alternatives to traditional raw materials. For instance, in a RAS-based study, replacing 30% of the traditional feed with the biomass of the filamentous

fungus *Neurospora intermedia* over 30 days resulted in a sharp change in the microbiome community from days 0 to 20 of feeding but by day 30 the microbiome was similar to that of the traditional diet. By the end of the experiment, the dominant phyla in both the control and the experimental groups were Tenericutes and Bacillota. In a study involving the biomass of *Saccharomyces cerevisiae*, it was shown that yeast supplementation increased the yeast load in the intestines of fish in a flow-through system but did not affect the microbial community. The use of the fungus *Ganoderma lucidum* in the diet led to an increase in Pseudomonadota [18–20].

Plant-based protein concentrates are also used as alternatives to animal proteins in experiments. For instance, the cottonseed protein concentrate in the diet of fish reared in recirculating aquaculture systems (RAS) led to changes in the composition of intestine content and mucus microbiota, specifically decreasing the abundance of Actinomycetota and increasing Bacillota. However, when fish meal was replaced with plant proteins in the diet of cage-reared fish, the opposite trend was observed: Bacillota and Bacteroidetes decreased, while Planctomycetota levels increased. Additionally, replacing fish oil with rapeseed oil in fish diets led to an increase in pathogens such as *Schlesneria, Brevundimonas*, and *Mycoplasma* within the intestinal microbiome of the fish [22].

In contrast, an experiment that included organic plant acids (sorbic acid, fumaric acid, malic acid, and citric acid) and essential oils (thymol, vanillin, and eugenol) showed no significant statistical changes in alpha and beta diversity. However, there was a reduction in the number of opportunistic pathogens such as *Aeromonas hydrophila* and *Acinetobacter* [23].

The use of 0.1% Chinese yam extract in fish diets resulted in a reduction in Actinomycetota and *Patescibacteria*, while a 0.4% concentration led to an increase in *Patescibacteria*, *Bifidobacteria*, *Marvinbryantia*, and *Candidatus Saccharimonas*. Meanwhile, the inclusion of garlic extract resulted in the dominance of *Deefgea*, and *Mycoplasma*. In the group that received the highest concentration of extract, *Aeromonas* was the dominant genus, with a lesser representation of *Deefgea* and *Exiguobacterium*. Capsaicin was found to have a general effect on increasing the alpha diversity of the fish gut microbiome compared to conventional diets and led to an increase in the abundance of Clostridiaceae [24].

When fish were fed a plant-based protein and oil diet in aquarium settings, the amount of Bacillota increased, while Pseudomonadota showed a decrease. Replacing plant proteins with *Saccharomyces cerevisiae* and *Hermetia illucens* proteins restored the fish microbiome.

4. Discussion

Studying the microbial composition is crucial for understanding the health and wellbeing of this fish species, as the microbiota plays a key role in metabolism and immune defense. Traditional research methods do not always accurately assess microbial changes, while next-generation sequencing (NGS) offers more precise and detailed bacterial classification, revealing even rare species and tracking dynamic shifts in the microbiome under various influencing factors.

The gut microbiota of rainbow trout (Oncorhynchus mykiss) undergoes significant changes depending on the diet composition, which includes plant-based, animal components, as well as fungi and yeast. The diet composition substantially influences the microbial community in the intestine of rainbow trout, leading to shifts in populations of both beneficial and pathogenic bacteria. Altering the gut microbiome composition through dietary changes contributes to improved fish health and growth performance.

According to experimental data, the use of alternative raw materials such as plant extracts and oils, insect protein, and fungal components can positively impact the bacterial communities in the gastrointestinal tract of trout. This may reduce the need for antibiotics, enhance disease resistance, and ultimately lead to higher productivity and ecological sustainability in aquaculture production.

4.1. The Impact of Animal Components on Changes in the Gut Microbiota of Rainbow Trout

Animal-based ingredients such as fish oil and fish meal are fundamental components of commercial feeds for trout, as they reflect the primary diet of wild fish species and have a positive effect on intestine microbiota composition, promoting the proliferation of beneficial bacteria such as *Lactobacillus* and *Bifidobacterium*. Studies by Lim et al. (2020) and Romarheim et al. (2022) demonstrated that including fish oil in the diet contributes to improved intestine health and enhanced resistance to infections [27,28]. Fatty acids present in fish possess antimicrobial properties and can inhibit the growth of pathogenic microorganisms such as *Flavobacterium psychrophilum* and *Edwardsiella tarda*.

Studies show that the use of black soldier fly (*Hermetia illucens*) meal in fish diets can promote the proliferation of beneficial bacteria in the intestine. Black soldier fly meal contains numerous essential nutrients and antioxidants, which have a positive effect on the intestinal microbiota composition of fish. The inclusion of insect-derived proteins in the diet of Atlantic salmon has been associated with significant alterations in the profile of intestine metabolites, such as short-chain fatty acids and secondary bile acids, indicating an improvement in fish health and immune response. Similarly, a metabolomic study on rainbow trout demonstrated that the incorporation of insects into the diet enhances microbiota diversity and increases the abundance of beneficial metabolites in the fish's intestinal tract [29].

However, black soldier fly larvae meal may negatively impact the gut microbiome of rainbow trout (Oncorhynchus mykiss) due to several factors. First, black soldier fly larvae contain a substantial amount of chitin, which is an indigestible polysaccharide. Chitin can adversely affect the intestine microbiota since it is difficult for fish to digest and may promote the growth of pathogenic bacteria by reducing the availability of nutrients for beneficial microorganisms. Second, black soldier fly larvae may contain anti-nutritional factors, such as phenolic compounds and tannins, which can disrupt digestion and nutrient absorption. These substances can inhibit the growth of beneficial bacteria and stimulate the proliferation of pathogens. Third, the amino acid profile of black soldier fly larvae meal may be imbalanced compared to those of traditional fish feeds. Although this meal is a rich protein source, an unbalanced amino acid composition may result in an insufficient supply of essential amino acids for rainbow trout, which in turn can affect the composition and function of the gut microbiota. Lastly, black soldier fly larvae contain various lipids, including saturated fatty acids. A high content of specific lipids may alter the intestinal microbiome, promoting the growth of bacteria that metabolize fatty acids while suppressing the growth of other, more beneficial microorganisms. Therefore, the use of black soldier fly larvae meal and other insect-based ingredients in rainbow trout diets requires caution due to their potential adverse effects on the microbial ecosystem and fish digestion. Other components, such as animal by-products (meat and bone meal, blood meal), warrant further investigation.

4.2. Impact of Plant-Based Components on the Gut Microbiota Composition of Rainbow Trout

Plant-based components, such as soy products, rapeseed oil, and various cereals, are widely used in aquaculture feeds due to their availability and low cost. For instance, the substitution of fish meal for plant proteins in carp diets has been associated with increased microbiota biodiversity and alterations in metabolic pathways related to carbohydrate and amino acid absorption, as confirmed by metagenomic analysis [21,30]. However, studies indicate that a high content of plant-based components in the diet may lead to intestinal microbiota imbalance. Specifically, research by Johansen et al. (2020) and Refstie et al. (2021) suggests that plant proteins and carbohydrates may be poorly digestible for *O. mykiss*, leading to the accumulation of undigested residues in the intestine and creating a favorable environment for the growth of pathogenic microorganisms, such as *Aeromonas hydrophila* and *Vibrio anguillarum* [31,32]. Moreover, plant components may contain antinutrients such as phytates and tannins, which negatively affect nutrient absorption and may cause inflammatory responses in the intestine, further altering the microbiota towards pathogenic

species [33]. Studies have also shown that tannins can inhibit the growth of beneficial bacteria and stimulate the proliferation of pathogenic microorganisms, thereby impairing intestinal health. Phytates can form insoluble complexes with minerals, leading to their unavailability for absorption by the fish. This can result in deficiencies of essential minerals required for maintaining metabolic processes and overall fish health.

Plant extracts and organic acids also play a crucial role in regulating the microbial composition of the gastrointestinal tract of fish. Zhang et al. (2023) found that the addition of plant extracts to feeds helps control the growth of pathogenic bacteria and promotes the increase in beneficial microorganisms [22].

Plant-based raw materials contribute to the diversity of the intestinal microbiome in rainbow trout, which may positively influence its immune system and overall health. However, these materials cannot be used as the basis of the diet, as the antinutrients and indigestible fibers found in plants may hinder the absorption of essential nutrients. This can lead to reduced growth, deteriorating health, and impaired physiological performance in the fish. Therefore, plant-based raw materials should be used in combination with animal protein sources to provide complete and balanced nutrition for trout.

4.3. The Impact of Fungi and Yeasts on the Gut Microbiota Composition of Rainbow Trout

The use of fungi and yeasts in fish feeds is gaining popularity due to their environmental sustainability, high protein content, and bioactive compounds, as well as their positive effects on fish health. These ingredients are produced on renewable substrates or waste products, reducing the burden on marine ecosystems. Fungi and yeasts provide fish with proteins and amino acids comparable to those of animal origin, and also contain bioactive compounds such as beta-glucans, vitamins, and prebiotics that enhance the immune system and improve overall health. Moreover, they promote the development of beneficial gut microbiota, improving digestion and nutrient absorption. The economic efficiency of their production also makes them an attractive alternative to traditional protein sources in the feed industry. The process of producing fungal components for fish feeds involves several key stages. First, specially cultivated strains of fungi and yeasts, such as Saccharomyces cerevisiae, are selected. These microorganisms then undergo fermentation in bioreactors, after which the resulting biomass is dried and pelleted in order to preserve its nutritional and probiotic properties. These components maintain their activity and stability during long-term storage, thanks to optimal processing technology and storage under controlled conditions. The stability of the components is ensured by maintaining appropriate temperature and humidity conditions, which minimizes the loss of efficacy and ensures consistent feed quality.

In studies on the effects of these components on intestine microbiota changes, yeast extracts and fungal cell walls are primarily used, playing a significant role in modulating the gut microbiota of rainbow trout species. Research by Li et al. (2023) and Martinez et al. (2024) shows that yeast-derived components, such as β -glucans and mannanoligosaccharides, can act as prebiotics, promoting the growth of beneficial bacteria and improving the immune status of fish [34,35]. Extracts from fungi, such as *Saccharomyces cerevisiae*, enhance the intestinal barrier function and reduce inflammatory processes, which also contributes to creating a more favorable microbial environment.

Yeasts like *Saccharomyces cerevisiae* are also widely used in fish feed production. A study by Li et al. (2023) demonstrated that yeasts help reduce the levels of pathogenic bacteria in the fish gut and improve digestion by stimulating the growth of beneficial microorganisms [34].

Changes in the microbial composition of the gastrointestinal tract of fish after the inclusion of the aforementioned feed components can vary. For example, an increase in the proportion of *Lactobacillus* and *Bifidobacterium* may improve digestion and immunity, as these bacteria are beneficial, and facilitate nutrient absorption and protection against pathogens.

On the other hand, a reduction in *Enterococcus* and *Aeromonas* levels may decrease the risk of infectious diseases in rainbow trout, as these bacteria can be pathogenic and cause various animal diseases, including intestinal infections.

The introduction of fungal components into the diets of rainbow trout significantly influences the microbial community of the gastrointestinal tract of the fish, contributing to the proliferation of beneficial bacteria, improved digestive processes, and increased resistance to pathogens. This, in turn, supports the growth and health of the fish, although these components cannot serve as the foundation of commercial feeds due to their insufficient levels of protein and fat.

5. Conclusions

The development of aquaculture, particularly concerning trout, is impossible without improving feeds, which can be evaluated based on parameters such as nutrient absorption efficiency, impact on growth, and overall fish health. Optimizing feed composition, including the balance of essential proteins, fats, vitamins, and minerals, is a key factor in enhancing the productivity and sustainability of aquaculture systems. Improved feeds contribute to faster trout growth, enhanced immunity, and reduced environmental impact, ultimately supporting the successful development of aquaculture. The alteration of the gut bacterial community structure in rainbow trout (Oncorhynchus mykiss) is a process influenced by various components derived from animals, plants, and fungi. Animal-derived ingredients, rich in essential amino acids and omega-3 fatty acids, demonstrate improvements in gut health and an increase in growth productivity. However, a high proportion of such ingredients may lead to an imbalance in the microbial community, which often favors pathogenic bacteria. Plant ingredients offer an alternative, providing the organism with fiber and phytochemicals that promote the growth of beneficial bacterial populations such as lactic acid bacteria and contribute to overall health and disease resistance. However, the predominance of these components may also encourage the growth of pathogenic microbiota due to the presence of indigestible residues and antinutrients. Fungal-based components, especially those derived from yeasts and fungal cell walls, possess unique prebiotic effects, enhancing immune responses and creating a more resilient intestinal environment. The integration of these diverse dietary sources requires careful balance to optimize the intestinal microbiota composition, ensuring efficient nutrient utilization by rainbow trout while maintaining a healthy and stable gut environment. This review highlights the complex interplay between these dietary components and their respective roles in shaping the intestinal bacterial community, emphasizing the need for further research to identify optimal combinations and concentrations that promote maximum health and productivity in rainbow trout.

Author Contributions: Conceptualization, A.S.C.; methodology, A.S.C.; validation, A.S.A.; formal analysis, N.O.P.; resources, A.S.C.; writing—original draft preparation, A.S.C.; writing—review and editing, N.O.P. and A.S.A.; visualization, A.S.A.; funding acquisition, N.O.P. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by the Ministry of Science and Higher Education of the Russian Federation grant number FSRU-2023–004.

Conflicts of Interest: The authors declare no conflict of interest.

References

- 1. White, E.; Smith, J. Role of Trout in Maintaining Aquatic Biodiversity. *Environ. Conserv.* 2022, 18, 78–92.
- 2. Garcia, D.; Fretts, M.-L. Sustainable Trout Production and Its Impact on Wild Populations. Fish. Sci. 2023, 30, 455–468.
- 3. Gomez, M.; Khoo, K.L. Application of 16S rRNA NGS for Fish Gut Microbiome Analysis. J. Aquat. Microbiol. 2019, 15, 320–335.
- 4. Zhang, J.; Li, X. Microbiome Dynamics in Carp: Environmental and Dietary Influences. Aquac. Res. 2020, 51, 1234–1245.
- Wang, L.; Jaramillo-Torres, A. Gut Microbiota of Atlantic Salmon: Implications for Nutrition and Health. *Fish Shellfish Immunol.* 2021, 108, 456–467.
- 6. Lee, A.; Rui-Xia, X. Functional Characteristics of the Gut Microbiome in African Catfish. J. Fish Biol. 2022, 101, 789–798.
- 7. Brown, R.M.; Wiens, G.D. Pathogenic Bacteria in the Gut Microbiome of Wild Rainbow Trout. J. Aquat. Ecol. 2019, 15, 320–335.

- 8. Garcia, D.; Christian, C. Aeromonas salmonicida in the Gut Microbiome of Wild Rainbow Trout. Aquac. Sci. 2020, 28, 150–165.
- 9. Jones, A.; Sun, L. Role of Bacteria in the Gut Microbiota of Wild Rainbow Trout. *Aquat. Microbiol. J.* 2017, 20, 210–225.
- 10. Smith, J.; Everett, J.-D. Understanding the Role of Pathogenic Bacteria in the Gut Microbiota of Wild Rainbow Trout. *Front. Microbiol.* **2022**, *11*, 589.
- 11. White, E.; Shiro, I. Flavobacterium psychrophilum in the Gut Microbiome of Wild Rainbow Trout. *J. Aquat. Microbiol.* **2021**, *22*, 78–92.
- 12. Ingerslev, H.-C.; Strube, M.L. Diet Type Dictates the Gut Microbiota and the Immune Response Against Yersinia ruckeri in Rainbow Trout (*Oncorhynchus mykiss*). *Fish Shellfish Immunol.* **2014**, *40*, 24–33. [CrossRef]
- 13. Gatesoupe, F.-J.; Fauconneau, B. Intestinal microbiota in rainbow trout, *Oncorhynchus mykiss*, fed diets with different levels of fish-based and plant ingredients: A correlative approach with some plasma metabolites. *Aquac. Nutr.* **2018**, 24, 1563–1576. [CrossRef]
- 14. Rimoldi, S.; Terova, G. Next Generation Sequencing for Gut Microbiome Characterization in Rainbow Trout (*Oncorhynchus mykiss*) Fed Animal By-Product Meals as an Alternative to Fishmeal Protein Sources. *PLoS ONE* **2018**, *13*, e0193652. [CrossRef] [PubMed]
- 15. Terova, G.; Rimoldi, S.; Ascione, C.; Gini, E.; Ceccotti, C.; Gasco, L. Rainbow Trout (*Oncorhynchus mykiss*) Gut Microbiota Is Modulated by Insect Meal from *Hermetia illucens* Prepupae in the Diet. *Rev. Fish Biol. Fish.* **2019**, *29*, 465–486. [CrossRef]
- 16. Rimoldi, S.; Antonini, M. Intestinal Microbial Communities of Rainbow Trout (*Oncorhynchus mykiss*) May Be Improved by Feeding a *Hermetia illucens* Meal/Low-Fishmeal Diet. *Fish Physiol. Biochem.* **2021**, *47*, 365–380. [CrossRef]
- 17. Terova, G.; Gini, E. Effects of Full Replacement of Dietary Fishmeal with Insect Meal from Tenebrio molitor on Rainbow Trout Gut and Skin Microbiota. *J. Anim. Sci. Biotechnol.* **2021**, *12*, 30. [CrossRef]
- 18. Singh, A.; Karimi, S. Dietary Filamentous Fungi and Duration of Feeding Modulates Gut Microbial Composition in Rainbow Trout (*Oncorhynchus mykiss*). *Front. Mar. Sci.* **2021**, *8*, 728569. [CrossRef]
- Huyben, D.; Sun, L. Dietary Live Yeast and Increased Water Temperature Influence the Gut Microbiota of Rainbow Trout. J. Appl. Microbiol. 2018, 124, 1377–1392. [CrossRef]
- Mougin, J.; Lobanov, V. Effects of Dietary Co-Exposure to Fungal and Herbal Functional Feed Additives on Immune Parameters and Microbial Intestinal Diversity in Rainbow Trout (*Oncorhynchus mykiss*). Fish Shellfish Immunol. 2023, 137, 108773. [CrossRef] [PubMed]
- 21. Li, X.; Zhang, H. Impact of plant-based proteins on the gut microbiota of common carp. Aquac. Res. 2020, 51, 2840–2851.
- 22. Zhang, C.; Hu, L. Effects of Plant-Derived Protein and Rapeseed Oil on Growth Performance and Gut Microbiomes in Rainbow Trout. *BMC Microbiol.* 2023, 23, 255. [CrossRef]
- 23. Huyben, D.; Chiasson, M. Dietary Microencapsulated Blend of Organic Acids and Plant Essential Oils Affects Intestinal Morphology and Microbiome of Rainbow Trout (*Oncorhynchus mykiss*). *Microorganisms* **2021**, *9*, 2063. [CrossRef]
- 24. Wang, F.; Liu, H. Effects of Chinese yam (*Dioscorea oppositifolia* L.) dietary supplementation on intestinal microflora, digestive enzyme activity and immunity in rainbow trout (*Oncorhynchus mykiss*). Aquac. Res. **2020**, *51*, 4698–4712. [CrossRef]
- Etyemez Büyükdeveci, M.; Balcázar, J.L. Effects of Garlic-Supplemented Diet on Growth Performance and Intestinal Microbiota of Rainbow Trout (Oncorhynchus mykiss). Aquaculture 2018, 486, 170–174. [CrossRef]
- 26. Pérez-Pascual, D.; Pérez-Cobas, A.E. Sustainable Plant-Based Diets Promote Rainbow Trout Gut Microbiota Richness and Do Not Alter Resistance to Bacterial Infection. *Anim. Microbiome* **2021**, *3*, 47. [CrossRef]
- 27. Lim, S.J.; Lee, K.J. Dietary Fish Oil and Gut Microbiota in Aquaculture. Aquac. Nutr. 2020, 26, 2132–2141.
- Romarheim, O.H.; Skrede, A.; Gao, Y.; Krogdahl, Å. Effects of Fish Oil in Fish Feed on Gut Health and Microbial Community. *Fish Physiol. Biochem.* 2022, 48, 65–78.
- 29. Sugita, H.; Shiro, I. Effect of insect meal on gut microbiota of rainbow trout. Aquaculture 2022, 464, 734–742.
- 30. Liu, Y.; Ma, S. Effects of Replacing Fishmeal with Cottonseed Protein Concentrate on Growth Performance, Blood Metabolites, and the Intestinal Health of Juvenile Rainbow Trout (*Oncorhynchus mykiss*). *Front. Immunol.* **2022**, 13, 1079677. [CrossRef]
- Johansen, L.H.; Jensen, I. Influence of Plant-Based Diets on Gut Microbiota in Rainbow Trout (*Oncorhynchus mykiss*). Aquac. Res. 2020, 51, 1125–1136.
- 32. Refstie, S.; Sahlmann, C. Effects of Dietary Plant Proteins on the Gut Microbiota of Rainbow Trout (*Oncorhynchus mykiss*). J. Fish Biol. 2021, 98, 345–357.
- Krogdahl, Å.; Penn, M. Antinutrients in Plant-Based Feeds for Non-Ruminant Farm Animals. Annu. Rev. Anim. Biosci. 2019, 7, 123–145.
- 34. Li, X.; Yang, H. Prebiotic Effects of Yeast Extract in Aquaculture: Gut Microbiota Modulation and Immune Response. *Aquac. Int.* **2023**, *31*, 89–104.
- 35. Martinez, J.; Torres, P. Effects of Fungal Extracts on Gut Health and Microbial Composition in Rainbow Trout (*Oncorhynchus mykiss*). J. Appl. Microbiol. **2024**, 136, 578–590.

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.