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EXPLORING UNCONVENTIONAL PLANT-BASED INGREDIENTS AND THEIR INFLUENCE ON SUSTAINABLE TROUT FARMING PRACTICES

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Abstract

Although fishmeal is an excellent feed component for trout, there are concerns about its sustainability. Overfishing and the environmental impact of fish meal production have raised questions about its long-term availability. Consequently, there's a growing interest in finding alternative protein sources for fish feed, such as plant-based proteins. The exploration of non-conventional ingredients in trout feed stems from a pressing need to safeguard the environment, promote aquaculture sustainability, and enhance consumer safety. By ensuring the safety of food products, consumer trust in purchasing aquaculture-derived fish is likely to increase, potentially reducing reliance on wild-caught fish. Within this framework, a comprehensive literature review was conducted to identify novel ingredient sources suitable for trout feed. This involved examining the origins of new plant-based ingredients, assessing their physical, chemical, and microbiological characteristics, and evaluating their impact on trout feed production. The findings of this study can inform and enhance feed management strategies in aquaculture, aligning with sustainability objectives and efforts to boost productivity.

Key words: *aquaculture, alternative fish diets, go green, emerging feed resource, environmental issues.*

INTRODUCTION

To ensure the sustainability of animal and its long-term viability, it is important to discover novel feed resources with superior nutritional value and conversion efficiency. Simultaneously, it is important to enhance the quality of animal products and maximize the efficient utilization of land and water resources (Holman & Malau-Aduli, 2012).

In recent years, the fish industry emerged as one of the fastest-growing sectors in global food production. In 2022, global fisheries and aquaculture production hit an unprecedented high of 223.2 million tons, comprising 185.4 million tons of aquatic animals and 37.8 million tons of algae. Of the total aquatic animal production, 62 percent was sourced from marine areas (with 69 percent from capture fisheries and 31 percent from aquaculture), while 38 percent was from inland waters (84 percent from aquaculture and 16 percent from capture fisheries) (FAO, 2024). This expansion has resulted in an increasing demand for fishmeal, an essential ingredient widely used in aquaculture feeds (Habib et al., 2008; Macusi et al., 2023; Rosenau et al., 2023). Therefore, aquaculture has outpaced other sectors and stands as the main consumer of fishmeal (Bachis, 2022). Following declining catch rates of wild fish, environmental issues associated with fishing methods, and the excessive energy and water requirements in the production of aquatic feed, finding of sustainable alternatives for fish fodder has become necessary (Holman & Malau-Aduli, 2012; Zhang et al., 2020; Rosenau et al., 2023). According this target, the World Trade Organization, in 2022, in Geneva, obtained a historic agreement between 165 countries aimed at reducing fishing subsidies and reducing overfishing worldwide.

Aquaculture confronts a significant challenge due to the increasing worldwide need for seafood coupled with stagnation in fishery capture production (Holman & Malau-Aduli, 2012). Aquaculture production had exceeded capture fisheries production for all sectors except marine finfish, which remains dominated by capture fisheries (Mair et al., 2023). 89% of the total aquatic animal production was intended for human consumption, providing an average of 20.7 kg per person. The per capita apparent consumption of aquatic animal foods has steadily increased, rising from 9.1 kg in 1961 to 20.6 kg in 2021, at an average annual growth rate of 1.4 percent. This growth has been fuelled by increased supply, advancements in preservation and distribution technologies, evolving consumer preferences, and rising incomes (FAO, 2024).

The reliance of aquaculture on feed ingredient produced by cooking, pressing, drying, and grinding fish or fish waste, which is widely condemned for the negative ecological effect, presents a sustainability concern. To ensure the sustainable expansion of aquaculture, it is essential to integrate alternative protein and lipid sources. Given the rapidly rising prices of food sources that provide nutrients for fish, as well as increasing demand of sustainable fisheries, a pressing necessity emerge to identify substitutes to these ingredients in compound feeds used in aquaculture (Zhang et al., 2020).

Therefore, possible protein and lipid resources which can serve as alternatives for fish meal and oil are being sought in plants. These substitutions have shown promise, given their biochemical composition and bioavailability. Nevertheless, usage of plant-based oil does come with certain disadvantages, primarily related to adulteration of fat structure in the fish's muscular tissue, and secondly their extractability (Bell et al., 2001; Torstensen et al., 2005; Pettersson et al., 2009).

One of the most traded fish species is rainbow trout *Oncorhynchus mykiss*. Currently, trout farming is spread all over the world: North and South America, Oceania, Europe, Asia, and Africa. In 2020, the worldwide production of rainbow trout reached approximately 960 K tons, representing approximately 4% from the total amount of farmed fish (Rashidian et al., 2020; Vaclavik et al., 2020; FAO, 2022).

In the context of Blue Transformation and in alignment with Go Green principles, sustainable aquaculture can address some of today's most urgent challenges. This includes ensuring food security, reducing the strain on wild fish populations, and lowering the climate and environmental impact of our food system. Additionally, it can provide consumers a wider variety of healthy and sustainable food products.

The review presents a bibliographic analysis focused on exploring new alternative vegetable ingredients and protein sources for trout feed, aiming to address concerns about the sustainability of fishmeal.

The study aims to support several significant practical point of view:

1. Reducing dependence on animal protein. Using plants in trout feed can reduce the need for animal protein, which is often obtained from sources such as small wild-caught fish. This can contribute to the conservation of natural resources and the protection of marine ecosystems.
2. Increasing food sustainability. Partial or total replacement of conventional trout feed ingredients with plants can contribute to greater sustainability of the food chain. Growing plants can be less intensive and less polluting than raising animals for food.
3. Improve the nutritional quality of feed. Choosing plant-based ingredients can improve the nutritional profile of trout feed by providing beneficial substances such as antioxidants, essential fatty acids and vitamins that can support fish health and well-being.
4. Reducing Ecological Footprint. Growing plants for food is often less harmful to the environment than raising animals. Reducing the ecological footprint of aquaculture through the use of plants can help maintain the health of aquatic ecosystems and conserve biodiversity.
5. Promoting innovation and research in the aquaculture industry. The study and implementation of non-conventional plant-based ingredients in trout feed can stimulate innovation in the aquaculture industry. This can open up new opportunities for research and development of more sustainable and healthier food products.

This study not only brings practical benefits, but can facilitate the transition to a more ecological and sustainable aquaculture model, contributing to protecting the environment and ensuring a sustainable food resource for the future.

MATERIALS AND METHODS

The review conducts an extensive examination of prior research concerning alternative feed constituents for trout, aiming to establish a comprehensive foundation for future investigations and experiments focused on substituting conventional feed components with non-traditional plant-based alternatives.

The study catalogued and analysed these alternative ingredients, mapping their geographic origins and conducting detailed assessments of their physical, chemical, and microbiological attributes.

Moreover, the study scrutinized the production implications associated with integrating these alternative plant-based constituents into trout feed formulations.

It explored the nuanced advantages and potential disadvantages of various unconventional feed sources, underscoring the urgent necessity for expanded research and development efforts in this field. The overarching goal is to ensure the sustainability and responsible advancement of aquaculture practices.

The research methodology involved leveraging a range of intensive research databases, including Web of Science, Google Scholar, PubMed, Scopus, and Science Direct.

These platforms were pivotal in gathering a comprehensive array of relevant papers covering diverse aspects of the environmental, economic, and nutritional impacts associated with alternative feed ingredients for trout.

Additionally, to enrich the literature search process, consultations were conducted with fisheries and aquaculture scientists, providing valuable insights and expert perspectives.

This holistic approach not only consolidates existing knowledge but also lays the groundwork for future studies aimed at optimizing feed formulations for trout farming.

By critically evaluating and integrating findings from multiple sources, this paper contributes significantly to the ongoing discourse on enhancing the sustainability and efficiency of aquaculture practices worldwide.

RESULTS AND DISCUSSIONS

New “green” ingredients

Developing plant-based ingredients for trout feed is a dynamic and expanding area of research and innovation within aquaculture. These ingredients are increasingly sought after due to their potential to mitigate the reliance on traditional marine-based components such as fishmeal and fish oil. The overuse of these marine resources has raised significant concerns about overfishing, which threatens marine biodiversity and disrupts ecological balance.

Plant-based ingredients offer a sustainable alternative that can help preserve marine ecosystems. They include a variety of sources such as microalgae and seeds from various plant species, each providing unique nutritional benefits. These ingredients are rich in essential nutrients like proteins, amino acids, vitamins, and fatty acids necessary for the healthy growth and development of trout.

Moreover, the use of plant-based ingredients can contribute to reducing the carbon footprint of aquaculture operations. Plant cultivation typically results in lower greenhouse gas emissions compared to fishing and fish processing. Additionally, the shift towards plant-based feeds can spur agricultural innovation, leading to more sustainable farming practices and the development of crops specifically tailored for aquaculture feed.

Incorporating plant-based ingredients into trout feed also addresses consumer demand for more sustainable and ethically produced seafood. With growing awareness about environmental issues and sustainability, consumers are increasingly favouring products that are not only healthy but also environmentally responsible. Aquaculture producers who adopt plant-based feed ingredients can tap into this market trend, enhancing their brand image and marketability.

Furthermore, ongoing research is focused on overcoming challenges associated with plant-based feeds, such as digestibility and palatability for trout. Advances in food technology and nutritional science are leading to the development of feed formulations that optimize the benefits of plant-based ingredients while ensuring the health and growth performance of the fish (Ivan et al., 2022).

The development of plant-based ingredients for trout feed is a key for the sustainability and future of aquaculture. It helps reduce environmental impact, supports marine conservation, meets consumer demands for sustainable products, and drives agricultural and nutritional innovation.

The main vegetable sources used in trout feed are algae (*Spirulina* sp., *Chlorella vulgaris, Tisochrysis lutea, Tetraselmis suecica, Nannochloropsis* sp.) and seeds rich in oils and protein from the plant species of the Brassicaceae Family (*Brassica napus* - rape, *Brassica carinata* - ethiopian mustard, *Camelina sativa* - false flax), sunflower (*Helianthus annuus*), lin (*Linum usitatissimum*), coconut (*Cocos nucifera*), and cotton (*Gossypium arboreum*).

It is important to note that the formulation of trout feed with plant-based ingredients should be carefully balanced to meet the nutritional requirements of trout. Additionally, ongoing research is essential to determine the optimal combination of plant-based ingredients and to assess their impact on trout growth, health, and product quality. Researchers and aquaculture companies are continually working to develop and optimize plant-based trout feed formulations to make trout farming more sustainable and environmentally friendly.

Areas of origin for vegetal ingredients

Several nations produce vegetable ingredients for fish feed. To comprehend the production of diverse plant-based components in fish farming and aquaculture, a distribution map was crafted. It delineates the principal countries engaged in production, organized in descending order according to productivity. The data utilized is derived from FAO statistics for the year 2022 (Figure 1) (FAO, 2023).

Figure 1. Different ingredients production used for Fishery and Aquaculture (original, using Google Maps) (https://www.google.com/maps/d/edit?mid=1NSaiLeO-DQkj6LQwxIAZBBVtCmv8CTk&usp=sharing)

Algae are considered a greatly appreciated type of protein, as well as essential amino acids (Fabregas et al., 1985; Becker, 1993) and vitamins (Becker, 2004). Global algae production (including cultivation and wild collection) increased over 3 times from 11.8 million (wet) tons in 2000 to 35.82 million tons in 2019; nearly all the growth was contributed by cultivation, while wild collection was relatively constant (Figure 2).

In 2022, the global algae production surged to 37.8 million tons (wet weight) from aquaculture, and 1.3 million from wild collection (FAO, 2024).

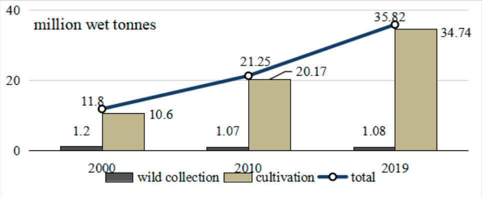

Figure 2. Global algae production, 2000-2019 period (based on data sourced from FAO, 2021)

In 2019, 35.8 million tons of world algae (including seaweeds and microalgae) production were contributed by 54 countries/territories with 97 percent of the production coming from cultivation. From total world production of algae (including seaweeds and microalgae of aquaculture and wild collection), the highest percentage from the total world production and the use of this production in aquaculture is in Asia (FAO, 2021) (Table 1).

Table 1. Global algae (including seaweeds and microalgae) production, in 2019 (based on data sourced from FAO, 2021)

Area/ Country	Total production (tons)	Share of world total (%)	Aquaculture share in total production (%)
World	35 818 961	100.00	96.98
Asia	34 881 600	97.38	99.10
China	20 351 442	56.82	99.14
Indonesia	9 962 900	27.81	99.55
Japan	412 300	1.15	83.80
Americas	488 144	1.36	4.87
Chile	427 508	1.19	5.28
Peru	36 348	0.10	0.00
Canada	12 655	0.04	0.00
Europe	287 386	0.80	3.99
France	51 683	0.14	0.74
Norway	163 197	0.46	0.07
Russia	19 544	0.05	54.10
Africa	145 259	0.41	81.33
Tanzania	106 069	0.30	100.00

Arthrospira is a type of cyanobacteria that forms tubular, multicellular trichomes in an exposed leftward spiral. Two species, *A. platensis* and *A. maxima*, are used to produce a dietary supplement known as spirulina. Large-scale microalgae culture began in Japan in the 1960s with *Chlorella*, and then Spirulina in 1970 in Mexic. Spirulina now is cultivated in over 20 states, including Brazil, China, India, Peru, Spain, and the U.S.A. The amount of spirulina produced worldwide on an annual basis is estimated to be around 3,000 tons per year (Shimamatsu, 2004), but FAO FishStat data suggests that the industrial production of spirulina is much higher than the previous estimate of 3,000 tons per year (FAO, 2006).

Besides the nations reported in FishStat, there are other significant producers of spirulina such as the U.S.A. (3394 tons in 2019), Taiwan, and Thailand, which are not accounted for in the FishStat reports. Spirulina can serve as a cost-effective alternative to animal-based protein sources in aqua feeds, and can either partially supplement or completely replace them. Compared to animal-derived feed ingredients, spirulina is a relatively low-cost option.

Rape (*Brassica napus*), originated as a cultigen in Southern Europe, it is now a domesticated plant species grown on almost all continents. The cultivation of *B. napus* started 6000 years ago in India, and it subsequently extent to East Asia around 1st century AD (Snowdon et al., 2007). Currently, *B. napus* is ranked as the third major variety of plant oil globally, just behind soybean and palm oil. Additionally, it is also the second main protein alternative globally, following soy (Heuzé et al., 2020). In 2021, world rapeseed production was 73.95 million tons, from which 26.58 million tons were canola oil. Normally, the European Union is responsible for about a third of the world's rapeseed production, which amounts to 60 million tons annually (FAO, 2023).

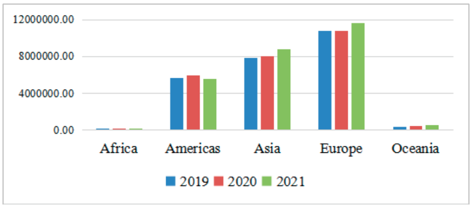

Figure 3. Production quantities of Rapeseed oil, crude (based on processed data from FAO, 2021)

In the European Union, Germany emerged as the leading producer with 3.505 million tons in

2021, contributing significantly to the total production. Beyond the EU borders, major producers include Canada, China, India, Australia, Russia, Ukraine, and the USA, while Africa and Oceania account for the lowest production levels (Figure 3) (FAO, 2023).

Ethiopian mustard (*Brassica carinata*), is a crop that is cultivated for its oilseed in Ethiopia. The culture of this plant was also introduced in other African countries, such as: Gabon, Ivory Coast, Kenya, Tanzania, and Uganda.

Lin or **flax** (*Linum usitatissimum*) is known as a plant rich in nutritional ingredients. It has been cultivated for thousands of years for germs, which is eaten whole, crushed, or pressed to produce flaxseed oil. Flax is taken into account for its possible welfares characteristics, for fuel or as fiber crop. The fat from seeds is so-called linseed oil and can be consumed as animal forage.

Sunflower (*Helianthus annuus*), which accounts for 8% of global oilseed production, is the fifth most cultivated oilseed crop worldwide. Originally native to Central North America, it has been widely distributed across the globe and is now found in regions ranging from Russia to South America.

In the realm of flaxseed productions obtained, Europe claims the top position, averaging 96.4% from 2019 to 2021. Oceania registers at 0%, while Asia, Africa, and the Americas collectively reach percentages of 3-4%. The largest producer in Europe is France with a production of 757680 tons. Also, the largest production of sunflowers worldwide is obtained in Europe (75%), and the largest producer in Europe is Romania (2,845,183.33 tonnes). More than 50% of the world's sunflower seed production is contributed by Ukraine, Russia and Argentina combined (FAO, 2023).

Coconut (*Cocos nucifera*), belongs to the *Arecaceae* Family and is the only surviving species in the Cocos genus. It is primarily found in the wet tropical biome and is omnipresent in warm areas by the seaside. It is considered a traditional image of the tropics. *C. nucifera* native range extends from Central Malesia to SE. Pacific and was originally farm by the indigenous ethnic minorities who lives on the islands in the Southeast Pacific and Indian oceans. The coconut tree was spread during the Neolithic period through seaborne migrations. The coconut tree was brought to various regions including India, Southeast Asia, Central Africa, Madagascar, and parts of North and Central America and South America. Majority of the worldwide coconut resource is now dominated by three countries, with the Philippines producing 42%, Indonesia producing 25%, and India producing 12%. In coconut oil production, Asia dominates with a productivity share ranging from 86% to 88% of the global production (Figure 4). The coconut tree is a versatile plant that has various uses: source of fuel, food and animal feed, medicinal plant and toxic substance.

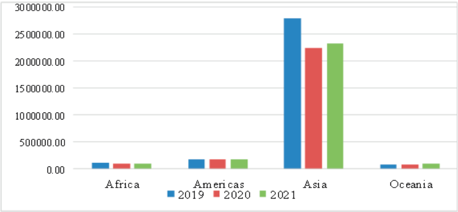

Figure 4. Production quantities of coconut oil (based on processed data from FAO, 2021)

Wild cotton species (*Gossypium* Genus) have been found in Mexico, Australia, and Africa, among other regions. Cotton plant was first domesticated in the Old World (likely in the Indus Valley of modern-day Pakistan) around 4500 BCE, and then independently in the New World (likely in present-day Peru) around 3600 BCE. There are about 50 Gossypium species and *Gossypium hirsutum* is the highly grown kind of cotton plant, representing almost the entire cotton crop in the world. *Gossypium barbadense* is the second most commonly grown species, representing around 2-3% of cotton global production. The remaining 1% is made up of *Gossypium arboreum* and *Gossypium herbaceum*, which are less widely cultivated (Chaudhry, 2010). Cotton is one of the primary natural fibers used by humans today, comprising approximately 80% of world natural fiber production. It is a major oilseed crop and a significant source of protein for animal feed where it is cultivated. Cotton is very important for agriculture, industry, and trade, particularly for tropical and subtropical countries in Africa, South America, and Asia.

As a result, the *Gossypium* genus has been a subject of interest for scientists for a long time.

The oil obtained from cotton seeds were a dietary staple in U.S.A. in the last hundred years, and prior to 1940s, it was the primary plant oil manufactured in this country. Currently, it is the third oil produced from plants in U.S.A., with a yearly average quantity of over 500,000 tons. *Gossypium* sp. oil represents up near 5-6% from whole home fat and oil source and is one of the most unsaturated oils, along with oils from seed plants as *Carthamus tinctorius, Zea mays, Glycine max*, *Brassica campestris*, and *Helianthus annuus*.

The largest cotton-producing region is Asia, followed by the Americas and Africa, Europe and Oceania registering the lowest productions (Figure 5) (FAO, 2023).

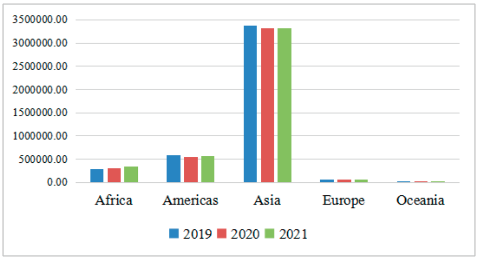

Figure 5. Production quantities of cotton oil (based on processed data from FAO, 2021)

Physical, chemical and microbiological characteristics of some plant-based ingredients

There are several species of **spirulina**, but the most commonly safe considered for consumption are *Arthrospira platensis* and *Arthrospira maxima*. Other species of spirulina include *Arthrospira fusiformis* and *Arthrospira biomass*, but these species are less commonly used for consumption. *Arthrospira* sp. is known for its high protein content. It typically contains around 50-70% protein by dry weight, more than other commonly used plant sources, which is way it is considered one of the most protein-rich food sources available (Phang et al., 2000). *Arthrospira* sp. is known to contain all essential amino acids, in a balanced composition, making it a complete protein source. Additionally, it contains high levels of fat molecule that contains two or more double bonds in their carbon chain (PUFAs) such as gamma-LA (GLA), which is a ω-6 fatty acid. *Arthrospira* sp. is also an abundant in vitamins, minerals, and photosynthetic pigments (Spolaore et al., 2006; Habib et al., 2008).

Spirulina is easily digestible due to the lack of cellulose in its cell walls, which is a unique benefit. Spirulina powder is a low-fat, low-calorie, and cholesterol-free source of protein, typically containing 60% protein, 20% carbohydrate, 5% fats, 7% minerals, and 3-6% moisture, making it nutritionally balanced.

Spirulina is a source of protein, with a high concentration fluctuating from 55% to 70% by dry weight, which exceeds most other commonly used plant sources. Its protein is complete and contains all essential amino acids, although with moderated levels of methionine, cysteine, and lysine equated with ordinary proteins found in products of animal origin. However, spirulina's protein profile is superior to other plant-based proteins such as those found in legumes. Spirulina is highly digestible because of the lack of cellulose in its cell walls, making it easy to assimilate.

Spirulina is rich in polyunsaturated fatty acids (PUFAs), comprising 1.5-2.0% of its total lipid content of 5-6%. Notably, spirulina is a good source of γ-linolenic acid (30-35% of total PUFAs), and also contains alpha-linolenic acid (ALA), linoleic acid (LA, 36% of total), stearidonic acid (SDA), eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA) and arachidonic acid (AA). Spirulina platensis is a promising source of γ-linolenic acid, which can be further increased by growing it under light-dark cycles either in a test centre or outside (Tanticharoen et al., 1994). Spirulina is abundant in a variety of minerals (potassium, calcium, chromium, copper, iron, magnesium, manganese, phosphorus, selenium, sodium, and zinc) and vitamins like thiamine (B1), riboflavin (B2), nicotinamide (B3), pyridoxine (B6), folic acid (B9), cyanocobalamin (B12), vitamin C, vitamin D, and vitamin E. Additionally, it contains high levels of β-carotene, which can be transformed to vitamin A. The presence of these vitamins, along with iron, potassium, and chlorophyll, can help promote the metabolism of carbohydrates, fats, and proteins, as well as support the growth and reproduction of skin, muscle, and mucosa.

Spirulina possesses the remarkable ability to detoxify and chelate toxic minerals. This means that it can neutralize or remove harmful substances from water and food, such as arsenic. Additionally, it has the potential to chelate or eliminate the poisonous effects of heavy metals found in water, food, and the environment.

Spirulina is rich in various pigments, including chlorophylla, xanthophyll, betacarotene, echinenone, myxoxanthophyll, zeaxanthin, canthaxanthin, diatoxanthin, 3-hydroxyechinenone, beta-cryptoxanthin, oscillaxanthin, as well as the phycobiliproteins c-phycocyanin and allophycocyanin.

The biological and chemical configuration of spirulina has been extensively studied, with analyses conducted on spirulina grown in different conditions, including laboratory settings, natural environments, and mass culture systems that use agroindustrial waste effluent. The composition of spirulina was set up to differ in feedback to the salt percentage of the growth environment. Additionally, detailed biochemical analyses have revealed the presence of various compounds and nutrients in spirulina, including protein, essential fatty acids, vitamins, and minerals. Vonshak et al. (1996), reported that the biochemical composition of salt-adapted cells differs from non-adapted cells, as they exhibit a decline in protein and chlorophyll and an upsurge in carbohydrate content.

Commercial production of spirulina is primarily based on superficial raceways where spirulina are agitated by a rotary vane. Nevertheless, there are still some instances where spirulina is commercially collected from natural environment.

When spirulina is cultivated in a laboratory setting, its productivity can be influenced by eight major environmental factors. These include luminosity (with a photo-period of 12/12 and 4 lumens), temperature (30°C), inoculation size, mixing rate, melted solids (ranging from 10 to 60 g/litre), pH (8.5–10.5), aquatic feature, and the existence of macro and micronutrients such as carbon, nitrogen, phosphorus, potassium, sulfur, magnesium, sodium, chlorine, calcium, iron, zinc, copper, nickel, cobalt, and selenium (Ciferri, 1983). Spirulina is a highly nutritious and sustainable food source that can be produced on a small scale with minimal resources, making it an attractive option for communities in need of nutritional support and economic development. Additionally, spirulina cultivation has the potential to mitigate environmental problems such as nutrient pollution and greenhouse gas emissions. Overall, the cultivation and consumption of spirulina has the potential to improve human health and well-being while promoting environmental sustainability (Habib et al., 2008).

Plant species of the Brassicaceae Family, like rape, have a primary purpose of cultivation for the oil-rich seed, which contains a significant amount of erucic acid (up to 50%) and glucosinolates. Erucic acid is known to cause damage to the heart muscles of animals, while glucosinolates have been found to interfere with iodine metabolism, leading to physiological disorders in the liver, kidneys, and thyroid glands. This interference can reduce growth and overall performance. The oils from the seeds of these species (camelina) are used in human and animal feed, as biodiesel for diesel engines and even as an aviation biofuel for jet aircraft engines (Resurreccion et al., 2021).

Rapeseed meal, or canola meal, is a by-product of the extraction of rapeseed oil from the seeds of *Brassica* species such as *Brassica napus, Brassica rapa, Brassica juncea*, and their hybrids. This residue is rich in protein and widely used as animal feed for various livestock. It is the second most commonly produced meal globally, after soybean meal (Heuzé, 2020). As fish meal supplies decline, vegetable-based proteins are being sought after by the aquaculture industry to provide the necessary amino acids for the high protein necessities of many farmed fish species, which are largely carnivorous. Canola meal has emerged as a significant ingredient in aquaculture diets across the globe, as it is able to fulfill this demand for vegetable-based protein. Although certain obstacles persist, several observations have demonstrated that canola meal can be an effective component in many fish food (Feed Industry Guide, 2019).

Ethiopian mustard seed contains great ranks of glucosinolates and erucic acid, which are considered undesirable for human and animal consumption. As a result, *B. napus* is preferred as an oilseed crop. However, the secondary product of *B. carinata* oil fabrication is consumed as protein meal as farm feed.

Linseeds and **linseed meal** gained significant interest after 1990 because of their great percentage of polyunsaturated fatty acids (PUFA), mainly alpha-linolenic acid (ALA) and conjugated linoleic acid (CLA). ALA is an omega-3 fatty acid, and linseeds and linseed meal are particularly rich in it (54% of the fatty acids). In addition to ALA, linseeds and linseed meal contain c18:1 (19%) and c18:2 (15%) fatty acids. Providing these fatty acids to livestock intakes it is changing the fatty acid balance of animal products in order to make more available one for consumers and for people wellbeing (Heuzé et al., 2018).

The great content of omega-3 fatty acids (ALA) in linseed meal causes a higher level of unsaturated fat in animal products, which leads to a smaller storing period. Additionally, the high omega-3 composition poses another difficulty since this fatty acid oxidizes quickly and becomes rotten, further reducing the storing interval.

Australia has developed cultivated varieties of *L. usitatissimum* that are specifically farmed to produce linseed oil with a reduced alpha-linolenic acid content. Linola was introduced in the 1990s as a low-linolenic acid variety of linseed, with reduced levels of omega-3 fatty acids. This made it a better option as fodder, as the high omega-3 content of linseed meal can cause a higher unsaturated fat content in animal products and shorten their storage time. However, linseeds and linseed meal still have some disadvantages. They contain a vitamin B6 (pyridoxine) antagonist, which may need additional supplementation. Additionally, linseeds contain 2-7% of mucilage (fibre), which are not processed by monogastric animals and may be harmful to young animals without treatments with enzymes (Heuzé et al., 2018).

Linseed meal is possible to be a potential protein supply in fish foods, but it has certain limitations because of its amino acid imbalance and antinutritional elements such as mucilages, tannins, phytates, and HCN. Feeding linseed meal in fish forages is restricted because of these factors, and protein digestibility which is generally low-slung, for instance, 70% in rainbow trout. However, these issues can be addressed through demucilagination, fermentation, and amino acid supplementation. By treating linseed meal, it can be administered to substitute 25 to 75% of the intake in fish feeds.

In rainbow trout, the digestibility of energy and protein from linseed meal is generally lower compared to other oil meals, with an average of 34% for energy and 70% for protein. In comparison, soybean meal has higher digestibility values of 77% and 89%, respectively, for energy and protein. Additionally, linseed meal has lower availability of essential amino acids such as histidine, valine, isoleucine, and lysine, which can limit its use as a protein source in fish diets (Gaylord et al., 2008; Gaylord et al., 2010).

Sunflower seeds. The original sunflower oil, also known as linoleic sunflower oil, has a high content of polyunsaturated fatty acids, particularly linoleic acid, which makes up about 68% of its fatty acid profile. It is also low in saturated fats, such as palmitic and stearic acid. Nevertheless, new hybrid varieties of sunflowers have been bred to adjust the fatty acid outline for specific purposes, such as high oleic sunflower oil, which has an upper concentration of monounsaturated fats and poorer values of polyunsaturated fats, making it more stable for high-heat cooking applications.

Sunflower seeds are grown primarily for their oil, but the by-product of oil extraction, sunflower meal, is also an appreciated component in livestock feeds due to its high protein content. Sunflower meal is commonly used in the diets of poultry, swine, and dairy and beef cattle as a source of essential amino acids and protein. Sunflower protein contains less lysine (around 4% protein) compared to soybean protein but has relatively higher levels of sulfur-containing amino acids, such as cystine and methionine (1.9% and 2.2% protein, respectively). Unlike other major oilseeds like soybeans, cottonseeds, and rapeseeds, sunflower seeds do not contain antinutritional factors, making it a safe feed for all livestock species. However, there may be concerns about residues and contamination (such as pesticides, insecticides, and mycotoxins like aflatoxin B1 and ochratoxin) during sunflower cultivation, harvest, and post-harvest operations (Heuzé et al., 2015).

Coconut oil has a natural flavour and aroma characteristic of coconut, and contains only a minor quantity of unsaponifiable substance along with trace amounts of tocopherols, tocotrienols, and phytosterols. It is colourless and composed of 92% saturated fatty acids, primarily triglycerides. Around 8% of the fatty acids are monounsaturated and polyunsaturated fatty acid. Most of the saturated fatty acids in coconut oil (nearly 70 percent) are medium-chain fatty acids (MCFAs). Medium chain fatty acids (MCFAs) are not commonly found in other plant oils, and coconut oil is unique in that it contains a high proportion of lauric acid (C12:0), ranging from 50-60%. Different portions of coconut oil also contain medium chain triglycerides. Because of its high MCFA content, the metabolism of coconut oil is different from that of other vegetable oils that mainly contain long chain fatty acids. Therefore, it cannot be assumed that coconut oil has properties similar to those of oils or fats that are primarily composed of long chain saturated fatty acids (92%). Coconut oil is reputed to have antibiotics effects, as well as exceptional medicinal qualities (Gopala et al., 2010; Perera, 2016).

Cottonseed oil is a type of herb oil that has an upper quantity of polyunsaturated fatty acids associated to saturated fatty acids, with a ratio of 2:1. The majority of its fatty acid profile is made up of 70% unsaturated fatty acids, which includes 18% monounsaturated (such as oleic acid), 52% polyunsaturated (such as linoleic acid), and 26% saturated fatty acids (primarily palmitic and stearic acids). Due to the presence of oleic, palmitic, and stearic acids, cottonseed oil is often referred to as "naturally hydrogenated", making it a suitable frying oil that doesn't require additional processing or produce trans-fatty acids.

The cottonseed oil industry asserts that cottonseed oil requires less hydrogenation compared to other polyunsaturated oils to achieve similar results. Additionally, refined and deodorized cottonseed oil is considered to be one of the purest food products available. Despite being highly refined, it still maintains its nutritional quality (List & King, 2006).

Gossypol is a toxic, yellow, polyphenolic compound produced by cotton and other members of the order *Malvaceae*, such as okra, which facilitates natural insect resistance. In the cottonseed oil refining process, the refining, bleaching and deodorizing steps act to remove the gossypol level

Coconut oil is composed of a significant amount of glycerides with low chain fatty acids and exhibits high resistance to atmospheric oxidation. Its chemical properties are characterized by a low iodine value, high saponification value, and a high content of saturated fatty acids. Moreover, it remains in a liquid state at average room temperatures of 27°C.

Aside from fatty acid glycerides, natural fats also contain minor amounts of other substances. These unsaponifiable constituents are mainly sterols. Coconut oil, for example, contains small quantities of tocopherols and phytosterols as unsaponifiable components.

Effects of ingredients of vegetable origin use in trout feed

Since 1974, algae have been added to the list of healthy foods by the United Nations World Food Conference due to its nutritional properties.

Microalgae hold significant potential as a sustainable alternative to fishmeal and fish oil in aqua feeds. They can be cultivated using seawater or wastewater on arid, infertile land with minimal nutrient input, while achieving a net biomass production that surpasses any terrestrial plant or animal. Microalgae biomass can accumulate high levels of protein (40-70%) and lipids, which are essential for fish growth and development. Additionally, they contain numerous value-added components such as carbohydrates, vitamins, antioxidants, probiotics, carotenoids, and amino acids that enhance fish health and quality (Shahin et al., 2023).

Specifically, *A. platensis* is considered a superfood (Jung et al., 2019) due to its antiviral, antibacterial, antioxidant, anti-diabetic, anti-cancer, and anti-inflammatory properties. This microalga has been shown to enhance fish development (Roohani et al., 2019), stress tolerance, and resistance to hunger, making it an effective supplement in fish farming (Nandeesha et al., 1998; Kumar et al., 2022). As a result of its beneficial properties, *A. platensis* has been increasingly used in fish diets to improve fish welfare.

Algae feeding experiments in aquaculture have been conducted to evaluate their potential as protein sources and as additives in fish feed, as a complete replacement for fishmeal. The use of algae as an additive has been found to have a positive effect on fish, as it can lead to lower levels of cholesterol and fat, and improve lipid metabolism (Holman & Malau-Aduli, 2012).

According to an experiment conducted on 216 rainbow trout for 10 weeks, Iranian researchers have concluded that *A. platensis* (spirulina) can serve as an alternative natural source of carotenoids instead of synthetic astaxanthin in the diets of rainbow trout. The researchers found that the inclusion of 7.5% *S. platensis* was sufficient to ensure pigmentation without any negative impact on fish growth (Teimouri et al., 2013a).

One study aimed to assess the possible protective effects of *A. platensis* on rainbow trout specimens exposed to three different doses of the toxicant CdCl2. Cadmium is a highly toxic heavy metal that is widely distributed in the environment. Unlike essential heavy metals like iron and zinc, cadmium is a non-essential element that can accumulate in the body and cause harm even at very low concentrations. Due to its toxic nature, cadmium exposure is a major concern for human health and the environment. It can enter the food chain through contaminated soil, water, and air, and has been linked to various health problems including kidney damage, bone demineralization, and cancer (Cicik & Engin, 2005). The exposure of *O. mykiss* to CdCl2 resulted in alterations in serum and liver function biochemical parameters, reductions in antioxidant enzyme activities, and an increase in markers of oxidative stress. However, Banaee et al., in 2022, showed that dietary supplementation of *A. platensis* was effective in minimizing or eliminating the negative effects caused by the heavy metal. The inclusion of *A. platensis* in the fish diet normalized all altered serum and blood parameters induced by CdCl2 exposure and had a protective effect on oxidative stress markers. These findings suggest that supplementing the diet of farmed fish with *A. platensis* may enhance their stress tolerance, which could improve their wellbeing, quality, and yield in aquacultural production systems.

Roohani et al. (2019), developed a complete randomized experimental design to assess the effect of dietary spirulina inclusion in fish meal sparing (FMS) on slow-growing juvenile Caspian brown trout. Fishes fed spirulina diets, despite the fish's sensitivity to diet composition had meaningfully higher body mass gain and specific growth rates compared to those fed the control diet, as well as higher protein and lipid efficiency and lower feed conversion ratio. Whole-body composition analysis revealed higher protein and lower lipid content in fish fed spirulina diets, with the highest protein deposition and lowest lipid content reported in this group. Spirulina supplementation also resulted in higher levels of beneficial fatty acids in fish fillet and increased fillet and skin color parameters. The study concluded that spirulina treatment improved growth, carcass composition, and pigmentation in juvenile Caspian brown trout.

A. platensis has been increasingly used as a dietary supplement due to its protein content and positive contributions, such as reducing oxidative damage (Mahmoud et al., 2021). Sheikhzadeh et al. (2019) found that feeding *Oreochromis niloticus* with algae-based feed resulted in improved antioxidant biomarkers, particularly in the gills and liver. The aim of 2019 study, was to investigate the effects of *A. platensis* on various parameters in rainbow trout, including growth, fillet composition, and mucosal antioxidant activity in the intestine, skin, and gill. The administration of 2.5% spirulina significantly increased total antioxidant activity in all three mucosal tissues, while feeding total antioxidant activity in the bowels were higher when fishes received doubled spirulina quantity. Gene expression analysis showed that adding 2.5% and 5% spirulina in forage, can upgrade the physiology of intestinal and skin tissue, in terms of catalase, glutathione S-transferase gene expression, antioxidant parameters and glutathione peroxidase action.

In the diets of three species of salmonids (*Salvelinus fontinalis, O. mykiss, Salmo trutta fario*), the fishmeal-based feed was completely replaced with *A. platensis*. There were observed differences in spirulina acceptance and conversion among the species, with the experimental diets being well-accepted except for brown trout. A species-diet interaction was observed, resulting in a reduction in final body weight due to spirulina supplementation for brook and rainbow trout ($p < 0.05$). Furthermore, the feed conversion ratio increased to the same extent in the spirulina-fed fish ($p < 0.05$), indicating that both species had similar abilities to convert the spirulina diet (Rosenau et al., 2022). The study found that there are changes to the colour and fatty acid profile of the fish. The colour of the fillets became more yellow and red due to the pigments found in spirulina, and this change was observed in both raw and cooked fillets. Additionally, the fatty acid profile of the fish was altered, with an increase in saturated and monounsaturated fatty acids and a decrease in polyunsaturated fatty acids. However, the study also found that the complete replacement of fishmeal with spirulina led to a reduction in growth and a decrease in feed conversion efficiency. Overall, the study suggests that using spirulina as a complete replacement for fishmeal may have some negative impacts on production performance and product quality traits.

Spirulina is a viable replacement for fishmeal, but it changes the colour of the fillet to yellow. This should not be seen as a disadvantage, but rather an opportunity to increase the perceived quality of the product. Studies show that consumers prefer the yellow colour of fillets produced with spirulina, making it not only a sustainable source of protein for fish, but also a way to increase the value of trout fillets. Fillet colour is the second most important factor when searching for fish fillets, after freshness, but the country of origin is the most important factor affecting consumer preferences. It is unclear how consumers would perceive traditional red fillets produced by feeding astaxanthin compared to the yellow/orange fillets produced with spirulina (Habib et al., 2008).

Teimouri et al. (2013b) conducted an experiment to investigate the impact of algae powder on concentration of carotenoids in blood (BCC). The findings indicated increasing the levels of *S. platensis* in the diet resulted in a significant increase in BCC. Fish fed with over 7% *S. platensis* had a higher concentration of carotenoids in blood paralleled with other meals. Positive correlations appear between concentration of blood carotenoids and development and average daily growth. In terms of food conversion and BCC it was observed negative relationship. In the muscle regression analysis showed that blood carotenoid levels were positively related to their final levels. Also, when rainbow trout were fed with *S. platensis*, carotenoids concentration in blood was highly correlated with the colour of fillets. The study found no significant difference in fillet carotenoid content after two weeks of storage at 4°C. Additionally, the carotenoid content in fish fed *S. platensis* remained stable at 4°C and at least for three months at -20°C. However, after six months, a significant decrease was observed in frozen storage. All these results indicated that the value of carotenoids in the blood will influence the final colour of the trout fillets, and during the storage of these food products the carotenoids will have a more stable state if the fish are fed with *S. platensis* (Teimouri et al., 2013b). Later researches suggested that low-level spirulina supplementation can increase the amount of beneficial polyunsaturated fatty acids (PUFA) (Teimouri et al., 2015; Roohani et al., 2019). However, with higher exchange rates of fishmeal with spirulina, this effect was reversed, implying that there is a limit to the amount of spirulina supplementation that can be utilized to avoid undesired reduction of desirable PUFA (Jafari et al., 2014).

It is worth noting that the quantity of spirulina included in fish feed can vary depending on the specific product and the intended purpose. The optimal dosage of spirulina for trout is not well-established and may depend on factors such as the age and size of the fish, as well as the specific nutritional needs of the fish. Some studies suggest that algal biomasses may not be highly effective as substitutes for rainbow trout diets due to their limited use of vegetal nutrients. The current experiment confirms this trend, as rainbow trout fry fed with a feed containing more than 12.5% algal biomass showed lower growth performances (Dallaire et al., 2007).

Another species of unicellular microalgae that can be used as feed in fish food is *Nannochloropsis oceanica*. There are studies that try to find out what would be the advantages and disadvantages of its use in trout.

In 2020, Sarker et al. investigated the effects of incorporating a mixture of microalgae meal (*Schizochytrium limacinum* and *N. oceanica*) into the diet of saltwater-reared rainbow trout. Three experimental diets with varying levels of microalgae meal were tested over a 10-week period. The results showed that increasing the inclusion of microalgae meal had a negative impact on growth performance and feed conversion ratio. However, it also led to improved liver health. The study suggests that microalgae meal can be included in rainbow trout diets as a sustainable replacement for fish oil, but higher inclusion levels may negatively affect growth performance.

Although there are studies that show that digestibility is lower at Nanno, this disadvantage can be overcome through processing technologies, which overcome the complex structure of the cellulosic wall and the high fiber content (Sarker et al., 2020). Various processing techniques, including extrusion and enzymatic treatment, were assessed for the *N. oculata* co-product. Extrusion processing at a lower temperature (90°C) yielded superior outcomes in digestible protein and amino acids compared to high-temperature extrusion (127°C). *N. oculata* was utilized in its raw form, as well as after treatment with enzymes and exposure to extrusion processes. The obtained product showed a good potential for improving protein and energy digestibility and also for essential amino acids, and omega-3 polyunsaturated fatty acids (n-3 PUFA) (Sarker et al., 2023).

Rapeseed meal is a commonly used food with essential nutrients for many fish species, but its great fibber volume confines its nourishing importance for predatory fish (McCurdy & March, 1992; Burel et al., 2000; Shafaieipour et al., 2008; Kaiser et al., 2022). Still, as when rapeseed meal is included in fish diets at rates lower than 50%, the fibre content is unlikely to exceed 8% of the diet, which is unlikely to impair growth performance in fish (Hilton & Slinger, 1986). Fish species, particularly carp, have been observed to have better tolerance for glucosinolates compared to swine and poultry. In the case of trout, the recommended upper limit for glucosinolates in the diet is set at 1.4 µmol/g, indicating their sensitivity to higher levels. Consequently, including rapeseed meals with low glucosinolate content in the diet can be beneficial, with recommended inclusion rates ranging from 20% to 30%. This approach ensures adequate nutrition for fish while minimizing any potential negative effects of glucosinolates (Feed Industry Guide, 2019). The mixture of rapeseed and soybean is often used as an alternative to fish meal in fish diets because they are good sources of protein and can replace the essential amino acids found in fish meal. In addition, the use of plant protein reduces the cost of the diet and also eliminates the risk of dioxin and PCB contamination, which is a concern for many consumer (Hertrampf & Piedad-Pascual, 2000; Newkirk, 2009). Studies have shown that the digestible energy content of rapeseed meal is lower than that of soybean meal in salmonids, with values ranging from 9.6-11.5 MJ/kg as fed, while soybean feed has an estimated digestible energy content of around 13.0 MJ/kg in its as-fed form. This value indicates the amount of energy that can be efficiently utilized by animals during digestion and metabolism (Sauvant et al., 2004; National Research Council, 2011).

The digestibility of rapeseed protein is high, ranging from 83% to 99%, making it an excellent source of amino acids, particularly for Atlantic salmon. In comparison to other plant-based protein sources, rapeseed protein has the most favourable amino acid profile (Anderson et al., 1992). Although rapeseed has been widely studied in rainbow trout, it has usually been established to have a negative effect on performance (Hilton & Slinger 1986; Burel et al., 2000; De Francesco et al., 2004; Drew et al., 2005; Newkirk, 2009; Alami-Durante et al., 2010; Collins et al., 2013). A study conducted on juvenile rainbow trout showed that including rapeseed meal in their diet at levels of 10%, 20%, and 30% during a 9-week period, had negative repercussions on the hepatosomatic indices, growing rate, feed conversion percentage, and state of immunity of the fish (Hernández et al., 2013). While most studies have found that rapeseed meal has a negative impact on the growth and performance of rainbow trout, there are studies which shown different results. For instance, some studies found that rapeseed meal covering 25 µmol/g glucosinolates could be included in supplies for young rainbow trout at up to 30 percents, without affecting growth, feed intake or feed efficiency (Burel et al., 2001). Another study found that rapeseed meal could be incorporated at 17.5% of the diet's dry matter, in combination with soybean meal at 14.5%, to substitute 40% of the protein delivered by fish meal. However, overall, rapeseed meal is not a preferred protein source for rainbow trout diets due to its negative impact on growth and performance (Güroy et al., 2012).

Canola meal is commonly used in the diets of salmon and trout; however, its inclusion is restricted due to the high protein requirements of these fish and the presence of heat-stable anti-nutritional factors. To evaluate the impact of different inclusion rates of canola meal, a meta-analysis was conducted using data from 12 studies and 30 data points focusing on the effect of canola meal in rainbow trout diets. The findings revealed that including canola meal at rates of up to 20% did not have a significant effect on the growth rate of the fish. This suggests that moderate inclusion of canola meal in the diets of rainbow trout is well-tolerated and does not compromise their growth performance (Collins et al., 2013).

The demand for commercially reared fish and crustaceans has increased, leading to a deficiency of fish oil, which in the future is predicted to worsen. As a result, there has been a move towards substituting fish oil with vegetable oils, which has been well-documented and typically has minimal effects on fish growth performance (Turchini et al., 2013).

Commonly used vegetable oils in salmon and trout diets are canola oil and rapeseed oil, due to their unsaturated fatty acids levels, with omega-3 and omega-6 fatty acids. In particular, due to its low levels of the omega-6 fatty acid linoleic acid, which helps to maintain a natural omega-3:omega-6 ratio found in fish, canola oil, is highly wanted. Vegetable oils are used extensively as a replacement for fish oil in fish diets. It has been shown to have little impact on growth performance in fish. Fish oil can be replaced in diets for rainbow trout, with canola oil, up to 90%, without adverse consequence in growing and developing performances. Additionally, in fillets, the total omega 3: omega 6 proportion, showed only minimal changes. Canola oil is therefore a suitable replacement for fish oil in fish diet (Turchini et al., 2013; Masiha et al., 2015).

Studies have shown that replacing up to 100% of the supplemental lipid in rainbow trout diets with canola oil does not have a significant impact on fish performance. This indicates that canola oil can be used in aqua feeds, as a suitable alternative to fish oil, without affecting growth or feed efficiency (Karayücel & Dernekbaşi, 2010).

Rainbow trout fish fed with different oilseeds (*B. napus*) had the final weight and weight gain significantly lower comparing to those fed with the control diet ($P<0.05$). However, fish fed with *B. rapa* and *B. carinata* at a 15% inclusion level had similar final body weight and gain as the control. Overall, the study suggests that all three oilseeds have potential for use in rainbow trout feeds (Anderson et al., 2018). Rapeseed oil has no harmful impact on the growth of rainbow trout and Arctic char, and it does not significantly affect the total lipid content of their white muscle. However, in rainbow trout, different tissues composition in fatty acid is influenced by the regime, and the muscles were more affected than the liver. Triacylglycerols are more susceptible to changes than phospholipids. Lower levels of highly unsaturated fatty acids, such as eicosapentaenoic acid and docosahexaenoic acid were found in muscle and liver of rainbow trout fed with rapeseed oil have, resulting in an inferior n-3:n-6 proportion. On the other hand, in the muscle of rainbow trout, the content of E vitamin increases with higher levels of rapeseed oil in the food. Although rainbow trout prefer fish oil over vegetal oil, they do not show any preference among different levels of vegetable oil inclusion in their supply (Rosenau et al., 2023).

Carinata flour (*B. carinata*) replaced fishmeal with different amounts of at 50 g, 100 g, and 150 g levels in Kasiga & Brown study (2019).

The outcomes of the study exposed that there was no significant modification in body weight or visceral weight. A 2020 study investigated the use of carinata (*B. carinata*) and camelina (*C. sativa*) seeds to enhance the nutritional composition of Rainbow trout feed. However, the inclusion levels of raw carinata and carinata seed meals in animal diets are restricted by the presence of antinutrients, mainly glucosinolates (GLS), sinapine, and crude fiber (Kasiga et al., 2020).

The seeds were subjected to processes to improve the chemical composition by cold pressing, extrusion, solvent extraction and aerobic conversion and then administered in fish feed, where the palatability was evaluated, depending on the acceptability shown by the fish. Carinata meals were found to have a higher crude protein content compared to camelina meals and also, the fiber content was lower in the case of feed with carinata. Carinata is believed to enhance nutrient utilization and boost meal efficiency in fish, making it a favored choice among feed manufacturers over other options (Kasiga et al., 2020).

Masiha et al. (2013a) carried out a study to assess whether a source of dietary lipid can be changed with **flaxseed** oil (also known as linseed oil) for rainbow trout fingerlings (*O. mykiss*). The findings of the study suggested that the fingerlings could be successfully raised on diets where fish oil had been substituted with flaxseed oil, without any significant impact on fish performance.

Linseed oil is still under research due to the digestibility problems it may have. Yu et al., 2019, has concluded following a study from 2019 that the Manchurian trout may have the ability to synthesize LC-PUFAs from ALA, and an appropriate linseed oil in substitution of fishmeal oil (<75%) could improve both the lipid metabolism and the oxidation resistance.

In 2022, Dupont-Cyr et al. conducted a study to explore the potential of linseed oil as a complete substitute for fish oil in the diet of *Salvelinus alpinus*, *S. fontinalis*, and their reciprocal hybrids, all belonging to the Salmonidae family. The research aimed to assess the influence of dietary lipid source on muscle fatty acid composition, growth performance, and feed utilization across four experimental groups fed diets containing either 100% linseed oil or 100% fish oil. The study findings revealed that replacing fish oil with linseed oil did not significantly impact growth performance, feed utilization, or muscle lipid and protein content. However, the muscle fatty acid profile was notably influenced by the type of dietary lipid. Substituting fish oil with linseed oil resulted in a decrease in certain omega-3 fatty acids (20:5n-3 and 22:6n-3) and an increase in 18:3n-3. Even hybridization between the closely related species did not seem to affect the expression of key enzymes involved in highly unsaturated fatty acid (HUFA) biosynthesis. The study suggests that linseed oil can effectively replace fish oil in the diet of charr without adverse effects on growth, feed utilization, or muscle composition.

It is true that linseed oil does not improve the characteristics and composition of the meat, nor does it bring significant changes in fish growth. However, it can be considered when we want to replace fish oil with a sustainable ingredient.

Also, Masiha et al. (2013b) evaluated the appropriateness of **canola** (COD) and **flaxseed** oils (FxOD) as dietary lipid sources for rainbow trout fingerlings, significant differences in the fatty acid composition were observed among fish fed different lipid sources. The levels of linoleic acid and α-linolenic acid showed substantial increases in fish fed COD and FxOD, respectively. Interestingly, the study found that the concentration of α-linolenic acid in the muscle was lower compared to the diets, indicating a high degree of metabolism of this fatty acid through β-oxidation and/or desaturation and elongation processes in rainbow trout fingerlings. Despite a decrease in eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) levels in the fillet of fish fed canola and flaxseed oils, the trout fillets remained a relatively rich source of these fatty acids, meeting the recommended daily intake of 500 mg/day of EPA plus DHA set by the International Society for the Study of Fatty Acids and Lipids. The study found that the fatty acid composition of rainbow trout flesh was influenced by the dietary fatty acid composition, and that fingerlings could be successfully raised on diets in which fish oil was replaced with canola and flaxseed oils. Additionally, the study concluded that the replacement of fish oil with these vegetable oils did not negatively impact the growth performance of the rainbow trout fingerlings (Masiha et al., 2013b).

Egg quality and reproductive performance of rainbow trout can be affected by various dietary oils. Fish oil (FO), linseed oil (LO), sesame oil (SO) and a commercial trout diet (CD) were fed to broodfish weighing approximately 870g, for about 5 months prior to reproducing period. Similar growing rate was among all fish studied groups. There were no significant differences between groups related to egg size. Moreover, the inclusion of LO or SO in the diet did not affect absolute fecundity, relative fecundity, or gonadosomatic index. As dietary lipid sources, LO and SO, can be used for broodfish and egg value and reproductive indices will have no negative changes. Additionally, efficient bioconversion of 18C fatty acids to 20-22C fatty acids was found in this study (Yıldız et al., 2020). The growth performance of rainbow trout was not impacted by diet, and there were only slight variations in semen volume, pH, and density ($p > 0.05$). Also, foods did not influence sperm microscopic qualities. Between the tested groups, semen osmotic values were different, higher values appear in groups fed with SO and LO. Sperm fatty acid summary, shows evidence of de novo biosynthesis of eicosapentaenoic acid, docosahexaenoic acid, and arachidonic acid. Despite this, reproduction indices were not significantly affected by food composition, suggesting that male rainbow trout are able to synthesize highly unsaturated fatty acids (Yıldız et al., 2021).

In 2011, Simmons et al., examined the impact on the quality of brook trout fillets when fishes are fed with **flax** diet. The study observed the impact of food adjustment using flaxseed oil-enhanced feed. The trout received either feed with fish oil (CD) or flaxseed oil (Flax) and then the fishes were harvested. The fillets were analysed for their proximate composition and fatty acid profile. During the storage period, raw fillets quality indices were also measured. Among the groups there were no significant changes, but the Flax fillets had a higher total omega-3 fatty acid content ($P<0.05$). Overall, the study suggests that a Flax-enhanced supply could have a positive impact on the brook trout bred in farms.

Czech researchers conducted an experiment on rainbow trout by adding 2% and 5% individually or a 5% mixture of sunflower or linseed oil to their diet. The firmness and juiciness of the fish meat were not affected by the addition of oils, but the taste of the fish was significantly influenced. The presence of sunflower or linseed oil did not have a significant effect on sensory characteristics, except for the intensity of the fishy taste. Linseed oil, which is a good source of n-3 polyunsaturated fatty acids (PUFA), contains a higher content of n-3 PUFA than sunflower oil, which can cause an off-flavour. However, none of the trout fillets were rejected due to the off-flavour. Both concentrations of linseed oil decreased the intensity of fishy taste. Therefore, the study recommends the use of linseed oil for the partial replacement of fish oil in rainbow trout feed (Drobná et al., 2006).

The effects of dietary supplementation of linseed (L) or sunflower (S) oil at 2.5% or 5%, or a mixture (5%) of both oils (LS5), was investigated on rainbow trout (*O. mykiss*). After 75 days of feeding, nutrient content in filleted fish was analysed. Weight gains, dry matter, fat, crude protein, cholesterol, saturated and monounsaturated fatty acids, arachidonic, eicosapentaenoic, and docosahexaenoic acids were not significantly different among the groups. However, meat from fish fed L5, S5, and LS5 had higher levels of polyunsaturated fatty acids (PUFA) compared to controls ($P<0.05$-0.01). The meat from fish fed L had less linoleic acid and more alpha-linolenic acid ($P<0.01$) compared to S-fed fish. S-fed fish had significantly higher levels of n-6 PUFA in their meat than all other groups. The concentration of n-3 PUFA was significantly ($P<0.05$-0.01) higher in the L-fed group compared to the S-fed group. The n-3/n-6 PUFA ratio in meat was significantly ($P<0.01$) higher in the L5 group compared to all other groups (Zelenka et al., 2003).

Adding 5% linseed or sunflower oils to extruded feed for rainbow trout, raised in recirculating systems, had several positive effects. It decreased the feed conversion ratio by 5.00% and 2.84% in trout fed EL and ES, respectively, compared to the control group. The weight gain of fish fed EL and ES increased by 5.03% and 2.14%, respectively, compared to fish in the C group. The inclusion of the oils did not affect the survival rate of fish in any of the groups, and it improved nutrient metabolism in rainbow trout. The study indicated that linseed and sunflower oils can successfully supplement extruded feed and replace some of the fish oil in rainbow trout diets. These oils did not negatively affect growth or survival rates but did improve weight gain and decrease feed conversion ratio. Vegetable oils in the diet increased the content of the essential fatty acids linoleic (LA) and α-linolenic (ALA), which can improve the lipid profile of the meat and be beneficial to human health (Zheliazkov, 2014).

In 2006, Italian researchers conducted a study on rainbow trout, exploring the possibility of herring and cod liver oil switching with coconut oil in the diet. The study lasted 231 days and involved four diets with varying levels of coconut oil, ranging from 0 to 13%. However, the study did not find any significant changes in the carcass features or meat composition of the rainbow trout fed with the different diets (Ballestrazzi et al., 2006).

Copra meal (derived from coconut) can be used as a feed ingredient for fish, but it is not an optimal one. This is because it contains less protein than fish meal or soybean meal and is lacking in lysine and sulphur amino acids. Although it is rich in arginine, excessive intake of dietary arginine can lead to lysine deficiency in animals, hence it is necessary to supplement fish diets with additional lysine and methionine to ensure adequate nutrition when using copra meal (Newkirk, 2009; Tacon et al., 2009). Additionally, it is important to note that phytic acid, tannins, and non-starch polysaccharides may appear in coconut oil as antinutritional combinations. These compounds can bind to nutrients, making them less available to the fish and reducing the overall nutritional quality of the diet. Therefore, careful consideration should be given to the inclusion of copra meal in fish diets and appropriate measures should be taken to minimize the negative effects of antinutritional factors (Tacon et al., 2009). Due to its high crude fiber content, copra meal is not an ideal feed ingredient for aquatic feeds. It is more suitable for herbivorous and omnivorous fish, where it can be included at rates ranging from 5-15%. However, for carnivorous fish, copra meal is less valuable and should be included at lower rates, typically between 5-10% (Hertrampf & Piedad-Pascual, 2000).

Despite the high melting point and saturated fat content of copra oil, which suggest it may be more suitable for warm-blooded mammals, it has been found to be an effective fat source in compound diets for the first-feeding larvae of common carp (*Cyprinus carpio* L.), even though they are reared at low temperatures (Fontagné et al., 1999; Fontagné et al., 2000a; Fontagné et al., 2000b). Studies have shown that including copra oil in the diet of rainbow trout did not have any negative effects on their growth (Figueiredo-Silva et al., 2012). Moreover, there were no adverse effects on reproductive performance in rainbow trout when copra oil was included in their diet (Ballestrazzi et al., 2006).

Luo et al. (2014) conducted a study on rainbow trout to investigate the effects of dietary fat source and level on plasma parameters related to health status. The experimental design involved feeding the fish high-fat (21%) or low-fat diets (11%) containing either highly saturated fat (derived from coconut oil - CO) or highly unsaturated fat (derived from fish oil - FO) as the sole fat source. The survival rate of the fish remained 100% during the three-week feeding trial for all dietary treatments. Final fish body weight and relative weight gain were significantly higher in the FO-High group ($P<0.05$) compared to fish fed the CO-High diet, with no significant difference observed between the FO-Low and CO-Low groups. Feed intake was also improved in the FO group compared to the CO group. Fodder productivity was however, not the same in all tests ($P>0.05$). According to a 15-week feeding study, the addition of CO in the rainbow trout meal did not result in a reduction in food intake. This finding is different from what has been observed in terrestrial animals, where the rapid oxidation and low retention of C12 in CO have been attributed to its satiating effect. In contrast, rainbow trout were found to deposit a significant portion of C12 and elongate/desaturate it into longer-chain fatty acids rather than quickly oxidizing it. This difference in fatty acid metabolism may explain why MCTs (medium-chain triglycerides) fail to create a satiety effect in rainbow trout

(Figueiredo-Silva et al., 2012). The ability of rainbow trout to efficiently include and convert C12, instead of rapidly oxidize it, differs from observations in mammals and may be the reason for the lack of a satiating effect of CO in this fish species.

In an experimental study, Lee et al., 2002, examined the efficacy of combining three diverse sources of processed **cottonseed** meal (CM) in the diets for juvenile trout. The objective was to completely replace fish meal (FM) protein with CM. The diets consisted of a combination of vegetal proteins (CM and soybean meal) and animal by-product proteins. The findings indicated that FM could be completely replaced with CM at a minimum level of 15% (equivalent to 25% replacement of fish meal protein) without any significant differences in growth rate and feed utilization. This suggests that incorporating CM as a replacement for FM in trout diets is a viable option without compromising the fish's growth performance and feed efficiency. However, in the control group haematocrit ranks were considerably higher compared with the group fed with CM-containing diets. The origin of the CM was found to affect its nutritive values in juvenile trout. Additionally, in the faeces of CM from Tennessee and Arkansas were found upper concentrations of total gossypol compared to that from California. The study also showed that gossypol optical isomer selectively collected in the liver and bile, while equal proportions optical antipode was established to be in the entire organism and faeces. Approximately 35-50% of dietary gossypol can be assimilated by fish, depending on the CM source, and the absorbed gossypol was almost completely eliminated (Lee et al., 2002).

Under the specific experimental conditions of a water temperature of 13 ± 1°C, researchers observed that juvenile rainbow trout with an initial average body weight of approximately 5.8 g had a mean feed conversion ratio (FCR) of 1.1 when fed diets containing either 100% fish oil or vegetable oils (such as soybean or sunflower oil). This indicates that the conversion of feed to body weight gain was relatively efficient, with similar FCR values observed for both fish oil and vegetable oil diets (Şener and Yıldız, 2003). Complete or partial replacement of fish oil with cottonseed oil in diets for rainbow trout did not have a negative impact on growth and feed utilization. The composition of fish fillet and total lipid levels in the liver of the fish is significantly affected. The feed conversion ratio (FCR) ranged from 1.3 to 1.4, indicate similar feed efficiency. However, in fish fed the cottonseed oil diet the hepatosomatic index (HSI) and viscerosomatic index (VSI) were higher paralleled to other supplies. Fish oil substitution with cottonseed oil led to a reduction in total fatty acid (n-3 PUFA) ranks and an increase in total n-6 PUFA levels in the fish fillet. Overall, the study demonstrated that replacing 50% of fish oil with cottonseed oil is feasible for rainbow trout diets, without compromising growth performance or fillet composition (Güler & Yildiz, 2011).

CONCLUSIONS

Fish meal and fish oil can be substituted in the diets of many fish species as long as all essential nutrients it provides are supplemented from other sources. This requires ensuring that the diet includes adequate levels of proteins, amino acids, lipids, vitamins, and minerals essential for fish growth, health, and development. Various plant-based ingredients, along with other sources like insect meal and yeast, can be used to create a balanced diet. These alternatives can be formulated to meet the nutritional needs of fish, thereby maintaining optimal growth rates, feed conversion ratios, and overall fish health.

The use of plant-based ingredients can help to reduce the environmental impact and cost of trout farming while maintaining the nutritional quality and growth of the fish.

Among different plant-based sources, algae, including spirulina, have shown promising results as a dietary supplement for trout, providing a source of protein, essential amino acids, vitamins, minerals, and antioxidants. Spirulina supplementation has been shown to improve growth rate, feed utilization, and immune function in trout.

Other plant-based sources that have been investigated for use in trout feed include Brassica sp. (such as canola), cotton, coconut, flax, and sunflower. These ingredients can provide a source of protein, energy, and essential fatty acids. Studies have shown that these ingredients can be included in trout feed at various levels without adversely affecting growth or survival. In the researched bibliography, studies were found regarding other species of oleaginous plants, which can be used in fish feeding, such as canola oil, palm oil. It should also be taken into account the possibility of mixing different types of vegetable feed to partial or total replace fish oil. Overall, the use of plant-based ingredients in trout feed shows promise as a sustainable and cost-effective alternative to traditional animal-based ingredients. However, it's important to carefully evaluate the quality and nutritional composition of plant-based ingredients and to optimize their use in trout feed to ensure optimal growth and health of the fish. Further research is needed to determine the most effective formulas and possibilities to mix different types of vegetable origin ingredients, for trout feed.

To ensure the sustainability of trout farming and aquaculture in general, it is essential to consider the environmental impact and ethical aspects of fish meal production and explore innovative and sustainable alternatives in fish feed formulation.

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