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Fat Deposition in Atlantic Salmon (*Salmo salar*) and rainbow trout(*Oncorhynchus mykiss*) Fed with Poultry By-Products

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Master thesis in Aquatic Food Production, 60 credits

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Abstract

In response to sustainability challenges in aquaculture and global protein demands, this thesis investigates the utilization of poultry hydrolysate, a byproduct of the poultry industry, as an alternative protein source in the diets of Atlantic salmon (*Salmo salar*) and rainbow trout (*Oncorhynchus mykiss*). This study aimed to assess the dietary effects on Atlantic salmon and rainbow trout by feeding them either a diet with 8% poultry meal (considered the control feed) or a diet with 3% poultry meal and 5% hydrolyzed poultry inclusion (referred to as the test feed). The fish were raised in commercial fish farms situated on the west coast of Norway. Sampling occurred twice: first when the fish weighed between 1-2 kg (rainbow trout in March 2023, 32 fish, and Atlantic salmon in June 2023, 80 fish), and second when they weighed between 3-4 kg (rainbow trout in June 2023, 52 fish, and Atlantic salmon in December 2023, 76 fish). Specifically, the effects of incorporating chicken hydrolysate at a 5% inclusion level were evaluated, focusing on its impact on visual fat deposition around the viscera, the heart, and in the fillet (myocommata), and chemical composition in the fillet (crude fat and fatty acids) and heart (fatty acids).

The study found that poultry hydrolysate significantly influenced body composition in rainbow trout, slaughter yield (SY) was lower for the rainbow trout, indicating a higher mass of visceral fat in the rainbow trout (SY=84%) compared with Atlantic salmon (SY=87%). Although changes in fat scores were not statistically significant for either species, inter-species analysis showed that Atlantic salmon consistently displayed higher visceral fat score that increased over time. While myocommata width increased within species over time, it was not significantly affected by the diet and showed no correlation with gaping. Moreover, the analysis of fatty acids showed improved profiles in rainbow trout fillets, with significantly higher levels of EPA and DHA, unlike in Atlantic salmon, where these fatty acids were primarily deposited in the heart. These findings highlight the differential responses to dietary poultry hydrolysate between species and underscore the importance of developing tailored dietary strategies to optimize fish health and enhance product quality in aquaculture.

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1. Introduction

Aquaculture has emerged as the most common method to produce seafood around the world, having undergone significant expansion over the past few decades to become the major mode of production. The aquaculture industry occupies a prominent role across a variety of different sectors in terms of the contributions it makes to the generation of employment, the promotion of economic advancement, and the improvement of nutritional outcomes (Lindland et al., 2019). As articulated in existing literature, it is noteworthy that fish species, particularly salmon and trout, exhibit a notably efficient food conversion ratio, approximating 1.1.

Nevertheless, the predominant cost component in the cultivation of these species pertains to the formulation and provisioning of feed, constituting a substantial proportion, approximately 45%, of overall production expenditures (Fiskeridir, 2021). Given the substantial weight of feed expenditure within the cost structure, both food manufacturing enterprises and scientific investigators have increasingly turned their focus toward the development and implementation of sustainable and ecologically responsible production methodologies. This strategic shift is motivated by the dual objectives of cost reduction and mitigation of environmental hazards inherent in the aquaculture sector (Bell & Waagbø, 2008).

In the realm of high production costs and intense market competition, the discerning and deliberate selection of raw materials in food processing stands as a conspicuous imperative. This selection process assumes a pivotal role, given its direct influence over essential attributes of fish fillets, namely taste, color, flavor, texture, and nutritional composition, all of which ultimately exert a profound impact on the marketability of the final product (Albrektsen et al., 2022).

Within the diverse array of ingredients in salmon feed, protein emerges as one of the most significant cost factors, profoundly affecting both the quality of the salmon fillet and the overall production costs, while also necessitating a focus on sustainability (Albrektsen et al., 2022). Given the variety of sustainable alternatives to fish meal available, such as insect meal, microalgae, microbial ingredients, plant-based proteins, and animal by-products, there exists an opportunity to select an option that is both more sustainable and abundant, depending on regional and climatic conditions (Cadillo-Benalcazar et al., 2020). A considerable proportion of the industry has shifted towards using soybean meal as a substitute for fish meal. However, concerns regarding the sustainability and environmental impact of soybean meal production, including deforestation and habitat destruction, persist (Cadillo-Benalcazar et al., 2020).

Among the various alternatives, this study concentrates on poultry by-product meal which is rich in protein. It is obtained from the rendering of poultry by-products, including parts not consumed by humans such as heads, feet, and viscera (Hekmatpour et al., 2018). The rendering process, which includes cooking, drying, and pulverizing these by-products, results in a concentrated source of protein. This investigation aims to explore the viability of poultry by-product meal as a sustainable and efficient protein source in aquaculture feeds, underscoring the necessity to evaluate protein sources that minimize environmental impacts and support the sustainability of aquacultural practices (Hekmatpour et al., 2018).

Within the context of the present research endeavor, a comprehensive investigation into the repercussions of incorporating hydrolyzed chicken protein into the production process is the primary objective. Specifically, the focus is centered on elucidating the impact of this inclusion on the extent of fat absorption and its subsequent distribution within the various tissue components of the fish fillets under scrutiny (Albrektsen et al., 2022). It is expected that the consumption of poultry hydrolysate protein in the diets of Atlantic salmon and rainbow troutwill result in significant variations in growth rates, biometric characteristics, and patterns of fat accumulation between the two species, with one species potentially display a more determined reaction in comparison to the other. This is the hypothesis that applies to both species.

1. Inclusion of poultry hydrolysate in the diet affect the fat deposition patterns in both Atlantic salmon and rainbow trout.

2. There are species-specific responses to poultry hydrolysate supplementation, affecting biometric traits, fat deposition patterns, and fillet quality in Atlantic salmon and rainbow trout.

2. Theoretical background

2.1 Sustainable feed

The biggest single bottleneck to be addressed in the quest to achieve the ambitious goals set by the Norwegian government and meet the industry's demand for sustainable feed ingredients is the increase in the supply of sustainable raw materials for feed (Hurdalsplattformen, 2021). The government has established a comprehensive objective of sourcing all feed for farmed fish and livestock from sustainable origins, with the overarching aim of mitigating greenhouse gas emissions within the food systems. Factors such as population expansion heightened demands on land and resources, and increased volatility in supply chains can exert significant pressure on food security (Regjeringen, 2022).

The societal mandate concerning sustainable feed endeavors to foster the emergence of novel and forward-thinking solutions aimed at optimizing resource utilization. Simultaneously, this undertaking holds substantial relevance in alignment with Norway's multifaceted aspirations spanning climate preservation, environmental conservation, sustainable feed production, employment generation, and economic value enhancement (Regjeringen, 2022).

Sustainability in aquaculture feed can be realized through a variety of methods and strategies that emphasize ecological, societal, and financial sustainability. To enhance the sustainability of aquaculture feed, several approaches can be adopted, such as using alternative ingredients, promoting a circular economy, prioritizing local sourcing, improving feed conversion efficiency, implementing traceability and certification, and conducting environmental impact assessments (Albrektsen et al., 2022). By adopting these tactics and guidelines, the production of aquaculture feed can advance towards greater sustainability, thus supporting the broader sustainability goals of the aquaculture sector and minimizing its impact on the environment (Cadillo-Benalcazar et al., 2020).

2.2 Feed ingredients

In a comprehensive statistical analysis of Norwegian salmon feed components from 1990 to 2020, the data reveals a steadfast adherence to traditional ingredients, with novel components making up a scant 0.4% of the feed (Albrektsen et al., 2022). Predominantly consisting of fish meal, fish oil, various vegetable proteins and oils, and carbohydrates, alongside essential vitamins and minerals, the composition shows minimal deviation over the three decades (Naylor et al., 2009).

However, Norway's ambitious plan to amplify salmon production by the year 2030 necessitates a significant 56% increase in feed volume, from the current 1.8 million tonnes to a projected 2.8 million tonnes (Naylor et al., 2009).

This projected expansion faces a formidable obstacle, as estimations suggest that only 140 thousand tonnes of this growth can be fulfilled using domestically produced ingredients, underscoring a pronounced feed gap (Tacon & Metian, 2008). Addressing this gap, the Norwegian aquaculture industry is poised to enhance sustainability by boosting local production of feed materials, thereby minimizing the dependency on imports. This strategic shift is not only environmentally astute—aiming to mitigate pollution from transportation and lower carbon emissions—but it also promises to strengthen economic resilience (Fiskeri- og kystdepartementet, 2009). By nurturing the growth of local raw materials, the industry can reduce environmental impact, foster economic stability, and create job opportunities. This approach is vital in cutting down the overall costs of food production and propelling the aquaculture sector towards a more sustainable future (Bell & Waagbø, 2008). The challenge and opportunity lie in innovating and expanding the utilization of novel feed ingredients, which will be crucial in bridging the feed gap and meeting the soaring demand sustainably (Shepherd et al., 2017).

Sustainable feed ingredients for salmon include marine-based ingredients like fish meal and fish oil from responsibly managed fisheries and fish processing by-products (Myhre, 2022). Algal meal from microalgae offers an eco-friendly alternative to fish oil, supplying essential nutrients and reducing dependence on wild fish populations. Insect meal, derived from organic waste, serves as a sustainable protein source, minimizing the environmental footprint compared to traditional sources (Hua et al., 2019). Utilizing by-products from seafood processing, such as tunicate meal and other marine animal remnants, supports waste reduction and circular economy efforts. Similarly, terrestrial animal by-products from poultry and pork provide sustainable protein options, enhancing circular food production and resource efficiency (Boyd et al., 2022). Additionally, exploring novel ingredients like blue mussels and photoautotrophic microalgae can diversify feed sources and decrease reliance on conventional options. Integrating these sustainable ingredients into salmon feed formulations allows the aquaculture sector to improve feed production's environmental and social sustainability while ensuring salmon receive the necessary nutrition for optimal growth and health.

Utilizing by-products from food production in both marine and terrestrial sectors as a high-quality component in animal feed is not only environmentally responsible but also promotes greater circularity.

In the domain of future ingredients, it is possible to categorize them into separate groups, which include harvested novel marine (Mesopelagic fish, and krill) and plant-based ingredients (grass, and tree biomass), farmed organisms (Black soldier fly, seaweed, blue mussels, yeast, fungus, and heterotrophic microalgae), and underutilized resources (poultry, pork, Whitefish, pelagic fish).

2.2.1 Fish meal (FM)

Fish meal (FM), derived from marine fish such as anchovy, menhaden, herring, and capelin, has traditionally been the cornerstone protein source in aquaculture feeds, prized for its rich content of essential amino acids, n-3 highly unsaturated fatty acids (HUFA), minerals, and excellent taste (Cadillo-Benalcazar et al., 2020). It boasts high digestibility, which facilitates efficient feed conversion and growth rates, while its palatability ensures minimal feed wastage by encouraging fish to eat more. Moreover, fish meal can be sourced sustainably from well-managed fisheries or as by-products from fish processing, playing a role in the circular economy by making use of fishery waste. It also has the added benefit of improving fish health through enhanced immune function and disease resistance (Bell & Waagbø, 2008). However, the use of FM is not without drawbacks. The environmental impact is significant, as overreliance on fish meal can lead to overfishing and negatively affect marine ecosystems and biodiversity (Naylor et al., 2009). Furthermore, FM can be costly, a situation exacerbated when demand outstrips supply or when sourcing it sustainably becomes more challenging. The sustainability of FM is also questioned when it is produced through unsustainable fishing practices, potentially leading to fish stock depletion and ecosystem degradation (FAO, 2020). Additionally, some fish may exhibit allergies to components in FM, which can affect their growth performance adversely. With the growing need for sustainable aquaculture, there's a push toward reducing reliance on FM by exploring alternative protein sources, aiming for environmental and economic sustainability in the long term.Therefore, while FM offers considerable benefits in terms of nutrition, its sustainability, cost, and environmental ramifications require careful consideration, suggesting that a balanced approach to FM use, alongside alternative protein sources, is essential for promoting sustainable aquaculture practices (Tacon & Metian, 2008).

2.2.2 Soy protein concentrate (SPC)

The integration of soy protein concentrate (SPC) into salmonid diets as an alternative to fish meal is gaining recognition for its considerable benefits, such as improved nutritional quality, cost savings, and environmental sustainability (Pelletier et al., 2018). Its role as a fish meal alternative not only aids in enhancing the health and performance of these fish but also enhances the cost-effectiveness of aquafeed, especially since SPC is generally more affordable than conventional fish meal. Moreover, the shift towards SPC usage in salmonid feeds addresses the pressing concern of resource sustainability (Hodar et al., 2020). Considering fish meal as a limited resource, replacing it with SPC substantially decreases the aquaculture sector's dependence on marine resources, thereby reducing the strain on wild fish stocks and aiding in the preservation of the sector's environmental sustainability (Pelletier et al., 2018).

Nevertheless, despite the apparent advantages of incorporating SPC into salmonid feeds, it is vital to acknowledge the broader environmental ramifications of such a shift, especially the carbon emissions resulting from transporting SPC. Mainly produced in Brazil and South America, SPC's transportation to aquaculture facilities in Norway incurs significant CO_2 emissions due to the extensive logistics required.

This fact underscores the need for a thorough evaluation of the sustainability of SPC dependency, emphasizing the necessity to account for the lifecycle emissions of feed components in environmental sustainability analysis (FAO, 2022).

Considering these environmental considerations, it is imperative to explore alternative solutions or substitutes that could alleviate these concerns while preserving the nutritional and environmental benefits. The pursuit of local or regional protein sources to replace or supplement SPC in salmonid diets represents an essential move towards minimizing dependency on long-haul transport, which could lead to a more ecologically sustainable approach to aquafeed production. This strategy encompasses the exploration of locally available ingredients, the enhancement of feed technologies, and the adoption of greener transportation options to tackle the challenges posed by SPC transportation. Therefore, although SPC represents significant progress in diminishing the environmental footprint and reducing the reliance on scarce marine resources in the aquaculture industry, the journey toward truly sustainable feed solutions necessitates continuous innovation and a comprehensive evaluation of the environmental expenses associated with feed production and logistics (Pelletier et al., 2018).

2.2.3 Poultry By-product Meal (PBM)

The integration of poultry by-product meal (PBM) into aquaculture diets, particularly as a substitute for traditional fishmeal and soybean meal, is supported by both a compelling supply argument and the necessity for sustainable practice. The substantial global poultry industry data reveals the enormity of potential raw materials available for aquafeed production. In the year under review, the global poultry industry slaughtered an astounding 73.79 billion chickens. This statistic, notably, does not include turkeys and emphasizes the magnitude of chicken processing, with Norway alone accounting for 72.6 million metric tons (MT) of chicken slaughter in 2021 (FAO, 2023). Given this context, the poultry by-product, primarily derived from the carcasses representing roughly two-thirds of a chicken's total mass, emerges as a substantial resource (Jedrejek et al., 2016).

This vast repository of by-products presents a significant opportunity for the aquaculture industry, particularly for salmon feed production. The rendering process of chicken carcasses yields a rich source of protein and essential nutrients, making PBM a practical and sustainable feed option. Given the high volume of available poultry by-products, there is an ample supply to meet the protein requirements of farmed salmon, thus alleviating the pressure on fishmeal and soybean meal resources (Galkanda-Arachchige et al., 2020)

Moreover, the economic benefits of PBM are clear, given its lower cost and reliable availability, providing a cost-effective solution for aquaculture operations. Additionally, the environmental sustainability of utilizing by-products from the poultry industry, which would otherwise go to waste, cannot be overstated.

Adopting PBM in salmon diets also addresses concerns over the environmental impacts of traditional fishmeal and soybean meal, including overfishing and the carbon footprint associated with soy cultivation and transportation (Galkanda-Arachchige et al., 2020).

Crucially, the acceptance and palatability of PBM by salmon, alongside its nutrient profile and growth performance outcomes, are key factors in its successful incorporation into aquafeed. Scientific studies support the efficacy of PBM as a substitute, with findings indicating that diets including PBM can support the growth and health of salmonids comparably to traditional fishmeal-based diets (Steffens, 1994).

In synthesizing these points, it becomes evident that the substantial volume of raw material from the poultry industry, alongside the myriad benefits of PBM, fortify the case for its inclusion in aquaculture diets (Nengas et al., 1999). The strategy aligns with global sustainability goals and the need for efficient utilization of available resources, marking a

significant step forward in the evolution of aquaculture feed strategies.The thesis thus posits that the poultry by-product meal, when carefully evaluated for nutritional adequacy and optimized for species-specific dietary needs, represents a superior, sustainable alternative feed ingredient that holds the potential to revolutionize aquaculture feed practices (Galkanda-Arachchige et al., 2020).

2.2.4 Chicken by-Product hydrolysates (CBPH)

In the context of salmonid feed formulation, the integration of chicken by-product hydrolysates (CBPH) offers a nuanced approach to enhancing feed quality through the lens of nutritional science and feed technology. The robust protein, peptide, and amino acid composition of CBPH aligns with the dietary requisites of salmonids, known for their specific nutritional needs to sustain optimal growth, health, and physiological development (Fallah-Delavar & Farmani, 2018). This compatibility underscores the potential of CBPH to serve as a cornerstone ingredient in salmonid diets, promising to elevate the overall nutritional intake without necessitating the over-exploitation of traditional marine resources (Villamil et al., 2017).

Delving into the functional merits of CBPH, their exemplary solubility, coupled with superior water-absorption capabilities, plays a pivotal role in the manufacturing of feed pellets. These characteristics ensure that the feed not only remains palatable and digestible for the salmonids but also maintains its structural integrity in aquatic environments, minimizing nutrient leaching and contributing to more efficient feeding strategies (Albrektsen et al., 2022). The emulsifying and foaming abilities inherent to CBPH further accentuate their utility, facilitating the production of cohesive and stable feed formulations that are critical for the homogeneous distribution of nutrients and for enhancing the sensory appeal of the feed to salmonids (Fallah-Delavar & Farmani, 2018).

Moreover, the economic rationale for incorporating CBPH into salmonid feed pivots on the pragmatic use of by-products, channeling underutilized resources into the creation of value-added inputs for aquaculture feeds. This practice aligns with the broader objectives of feed cost optimization and resource efficiency within the aquaculture industry, presenting a viable alternative to more costly protein sources (Bell & Waagbø, 2008). The strategic use of CBPH can lead to a recalibration of feed formulation costs, offering a pathway to more economically sustainable aquaculture practices without compromising on the quality or nutritional integrity of the feed (Naylor. et al., 2009).

In the realm of health benefits, the antioxidant properties of CBPH and the presence of bioactive peptides offer intriguing prospects for salmonid welfare. These components may play a role in enhancing immune responses and in mitigating stress-induced physiological challenges, thus contributing to the overall vitality and resilience of salmonid populations within aquaculture systems (Lim et al., 2023).

The incorporation of CBPH into salmonid feed, therefore, extends beyond mere nutritional supplementation; it represents a thoughtful convergence of dietary science, feed technology, and economic pragmatism, aimed at bolstering the health and efficiency of salmonid aquaculture operations. This approach not only fulfills the immediate nutritional needs of salmonids but also contributes to the broader goals of innovation and sustainability in aquaculture feed design (Albrektsen et al., 2022).

Norway largely follows the regulations set by the European Union. In 2009, a regulation was introduced requiring that animal by-products be free from contamination and diseases before being used in animal feed intended for human consumption (Regulation, 2009).

Poultry by-products, in particular, are prone to contamination by various pathogenic bacteria such as salmonella, campylobacter, pseudomonas, serratia, staphylococcus, enterococcus, and listeria (Rouger et al., 2017). To reduce the risk of contamination, poultry by-products can be subjected to testing and sterilization processes. Additionally, hydrolysis offers a cost-effective method for further processing these by-products, facilitating their reuse in animal feed applications (Rouger et al., 2017).It must not be overlooked that market acceptance is restricted due to emotional and ideological factors, along with a lack of trust in food safety regulations. These issues significantly impact market adoption in Norway and Europe (NCE, 2022) .

2.4 Distribution of nutrients within fish

In aquatic nutrition, nutrient partitioning is a critical process that determines the distribution and utilization of fats, proteins, and sugars across a fish's organs, tissues, and various body regions. This systematic allocation effectively segregates the fish's structure into lean body mass (LBM) and specialized zones for fat storage (Jobling, 2001). The composition of LBM is consistently water and protein-dominant in fish species, regardless of their stage of development, with a modest presence of minerals, carbohydrates, and structural fats. During their growth phase, fish expand their LBM and simultaneously enhance their lipid storage capacity (Jobling, 2001).

The evolving chemical composition of fish throughout their growth signals the differing accumulation rates of LBM and lipid reserves. Under conditions of limited feed supply or during periods of starving, fish adapt by redistributing nutrients from different body parts to satisfy energy requirements, consequently altering their body composition (Jobling, 2001).

Seasonal fluctuations in body composition are particularly pronounced in fish from temperate or varied climate zones. Influenced by factors such as feed accessibility, growth phases, and reproductive cycles, these fish typically build up lipid reserves during periods of warmer weather, utilizing these stores for energy in colder seasons when feeding is reduced. Notably, these lipid reserves also play a vital role in reproductive organ development.

Fish species have distinct lipid storage patterns in their bodies, influenced by their type and diet. Studies of the lipid-moisture interplay in species such as the European eel, whitefish, sockeye salmon, rainbow trout, Atlantic salmon, brown trout, and sea bass have shed light on the effects of dietary composition on lipid accumulation (Jobling, 2001).

Moreover, farmed fish fed with uniform, nutrient-rich diets are particularly adept at forming substantial lipid reserves. The bodily location of these lipid deposits can affect multiple aspects of farmed fish, including yield losses during processing, fillet texture, preservation qualities, and the nutritional value of the fillet, underscoring the complex relationship between nutrient partitioning, fish development, and the resulting quality of aquaculture products (Jobling, 2001).

2.5 The Impact of diet on lipid deposition pattern

The texture, flavor, and color of fish fillets are significantly influenced by their fat content, which can vary based on factors such as species, diet, feeding frequency, and the season.

Fish skeletal muscle contains two primary lipid groups: triglycerides, which store energy in fat depots, and phospholipids, which are essential for cell membrane structure (Acharya, 2012). Fatty acids, the building blocks of these lipids, are classified by their saturation level into saturated, monounsaturated, and polyunsaturated fats. Notably, the presence of n-3 fatty acids, such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), has been associated with a reduced risk of cardiovascular diseases (Méndez et al., 1996).

The type and lipid composition of the diet ingested by salmonids are known to influence the fatty acid composition of their tissue. While the precise effects of the total amount of dietary lipids on fat distribution and sensory qualities are less clear, it has been found that increased fat content in the diet corresponds with enhanced juiciness in the fillets of rainbow trout

(Johansson et al., 1991). Furthermore, the diet's composition is crucial in determining the fish's body composition, affecting everything from fat and protein levels to moisture balance. Fish that are fed high-lipid diets have a greater propensity for fat storage, resulting in a higher body fat percentage compared to those on low-lipid diets. The dynamic nature of these fat stores is also influenced by the diet's specific fatty acid content, which has implications for the fish's overall physique (Bell et al., 2010).

Protein content within the fish is contingent upon the diet's composition as well, with dietary variations capable of altering the protein profile within the fish's body, which in turn affects body structure. The moisture content within fish tends to be inversely related to body fat; diets high in lipids raise the body fat content and, simultaneously, reduce moisture levels, further altering body composition (Jobling, 2001) .

Seasonal variations also affect the fish's body composition, as seen in the fluctuating lipid content in farmed Atlantic salmon, which peaks in the spring and early summer in conjunction with increased feeding and reproductive activities. This seasonal pattern suggests that the diet influences not just daily nutrient distribution but also plays a significant role in the patterns of lipid storage and utilization throughout the year (Mørkøre & Rørvik, 2001).

2.6 Flesh quality

Fish muscle myotomes are arranged in folded sheets connected by layers of connective tissue called myocommata along the fish's length (fig.1) (Jacobsen et al., 2019). Gaping occurs when this connective tissue (CT) weakens and fails to hold the muscle together, creating holes and slits in the fillet. This issue significantly impacts the aquaculture and fisheries industries because it makes fillets look unappealing and less suitable for specialized food production, leading to lower quality ratings and reduced pricing (Pittman et al., 2013). Fillets exhibiting gaping are compromised in strength, unable to hold the muscle blocks together, and may easily disintegrate. Moreover, gaping detracts from the fillet's visual appeal and can pose challenges for slicing equipment, often resulting in these fillets being devalued and sold at a lower price. Additionally, the unattractive appearance of gaping fillets often leads to consumer rejection (Jacobsen et al., 2017).

Several variables, including the size and age of the fish, stress levels, and protein content, can influence the fillet of the fish (Wang, 2016). Stress from handling prior to and during harvesting is a significant factor contributing to gaping and softer fillets. This stress can induce an acidic

environment within the muscle, increasing Cathepsin L activity, which in turn leads to the breakdown of collagen, softening the fillet texture (Jacobsen et al., 2017).
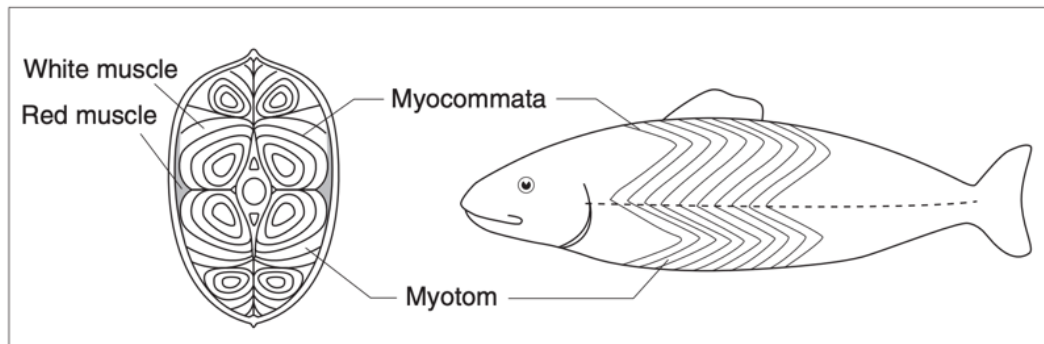
Fig.1, Fish with a cross-section displaying myotomes and myocommata, alongside white and red muscular tissue (Jacobsen et al., 2019)

Also the presence of melanin in fish fillets can significantly reduce product quality, affecting economic returns This issue is prevalent not only in salmon across the Norwegian coast but also in major salmon-farming countries like Chile, the UK, and Ireland. Reports indicate that this problem costs the Norwegian fish farming industry approximately one hundred million euros annually.Melanin spots on salmon fillets, which appear as light shades, dark spots, and grey or red spots, are caused by melanin accumulation and blood pigments from hemorrhages or scar tissue. These spots form due to inflammation and scar formation, indicating both acute and old tissue damage (Wang, 2016).

Melanin in fish fillets lowers product quality and economic value. This problem affects salmon in Norway and other countries like Chile, the UK, and Ireland. It costs Norway's fish farming industry about one hundred million euros each year. Melanin spots, seen as light or dark spots, and red or grey spots on fillets, result from melanin buildup and blood pigments from injuries. These spots indicate inflammation and old tissue damage (Wang, 2016).

3. Material and methods

3.1 Experimental setup

The fish used were Atlantic salmon (*Salmo salar*) and rainbow trout (*Oncorhynchus mykiss*), farmed at three different commercial locations: Garvik (rainbow trout), Litle Lunnøy (Atlantic salmon), and Krigsholmen (Atlantic salmon) (fig.2).

At each location, the fish were fed two different diets: a control feed and a test feed. The control feed contained 8% poultry meal, while the test feed had 3% poultry meal and 5% hydrolyzed poultry meal. The feeds were produced by Aller Aqua, Denmark.

The raw material composition for Atlantic salmon and rainbow trout is shown in Table 1 and Table 2, respectively. The chemical composition is in Table 3. Figure 3 provides an overview of the number of cages, the number of fish per cage, initial weight at sea transfer, and smolt producer.

Rainbow trout were sampled for analysis in March 2023 (first sampling) and June 2023 (second sampling) at Garvik. Atlantic salmon were sampled in June 2023 (first sampling) at Litle Lunnøy and Krigsholmen, and in December 2023 (second sampling) at Litle Lunnøy. At each sampling, eight fish were randomly selected from each cage (fig3).

Fig2. Maps of fish sampling sites at Garvika, Litile Lunnøy, and Krigsholmen in Norway, indicated by red pins. Source: Norwegian Mapping Authority (https://www.norgeskart.no).
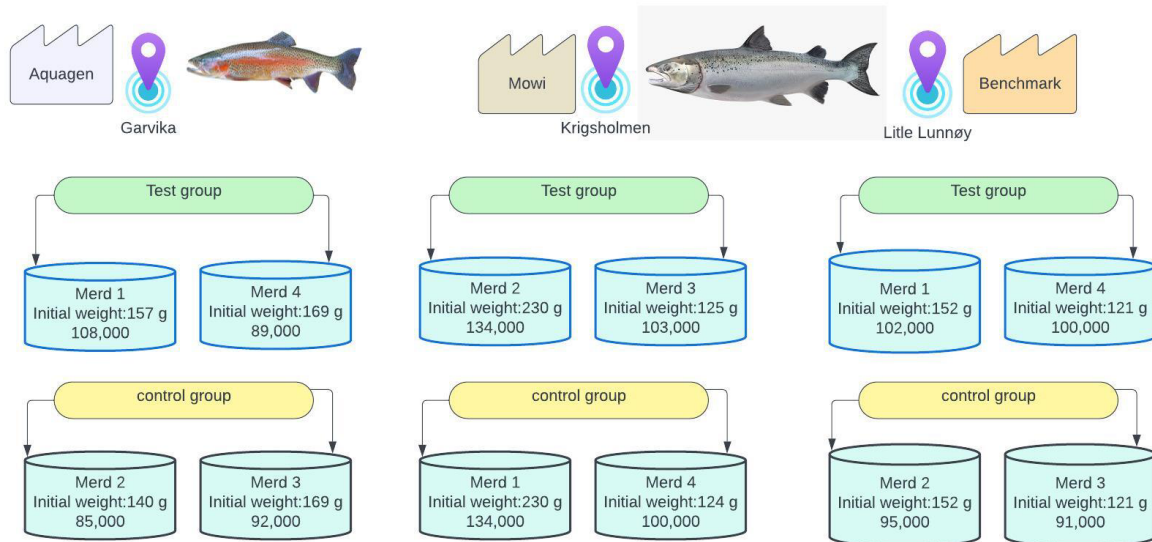
Fig3. Participating Suppliers and Research Locations: Aquagen, Mowi, and Benchmark respectively in Garvika, Krigsholmen, and Little Lunnøy. Each site hosted two control and two test groups for the study.

3.4 Fish feed

This study aimed to assess the dietary effects on fish populations by utilizing two distinct feeding strategies. The standard commercial feed (contained poultry meal), termed the control diet, served as the base line for comparison. In contrast, the test diet was modified by supplementing with 5% chicken hydrolysates, thereby enhancing the base commercial feed. Fish were maintained on either the control or test diet throughout the marine production cycle to ensure consistent feeding conditions.The feeds utilized for Atlantic salmon and rainbow trout, supplied by Aller Aqua, were documented in respective tables(table1, table2, table3). An inclusion of 5% chicken hydrolysates was a defining feature of the test diet. Alignment of the feed compositions with the specific dietary requirements of the studied fish species was meticulously planned. Diets supporting Atlantic salmon included protein-to-fat ratios of 44/28, 42/30, and 38/32, with corresponding pellet sizes of 3&4.5 mm, 6&9 mm, and 9 mm to cater to different growth stages. The formulated diets for rainbow trout adhered to protein-to-fat ratios of 44/28, 43/30, 38/32, and 37/34 with identical pellet sizes, ensuring that the nutritional needs of the species were met.

Table1, Composition of aquafeeds for Atlantic salmon, detailing the ingredients and their respective inclusion levels (expressed as a percentage of the total feed) for both control and test diets.

Atlantic salmon trials	Control				Test			
Pellet size	3 & 4.5 mm	6 mm	9 mm	9 mm	3 & 4.5 mm	6 mm	9 mm	9 mm
protein-to-fat ratios	44/28	42/30	38/32	37/34	44/28	42/30	38/32	37/34
Pea protein	0	0.81	6	6	0	0.81	6	6
Soy protein concentrate	10.72	12	10.49	8.86	10.72	12	10.49	8.86
Fishmeal LT	19.14	14.03	12.50	12.50	19.14	14.03	12.50	12.50
Fishmeal SA	10	10	10	10	10	10	10	10
Poultry meal	8	8	8	8	3	3	3	3
Hydrolysed poultry meal	0	0	0	0	5	5	5	5
Hydrolysed feather meal	6	6	6	6	6	6	6	6
Rapeseed oil	14.83	17.22	18.49	20.60	14.83	17.22	18.49	20.60
Fish oil	8	8	9	9	8	8	9	9
Emulsifier	0.30	0.40	0.50	0.50	0.30	0.40	0.50	0.50
Wheat	11	11	14.44	13.73	11	11	14.44	13.73
Corn gluten	8	8	0	0	8	8	0	0
Pea starch	1	1	1	1	1	1	1	1
Diamol	1	1	1	1	1	1	1	1
Monoammonium phosphate	1.06	1.06	0.86	0.91	1.06	1.06	0.86	0.91
L-Lysine	0.12	0.52	0.66	0.79	0.12	0.52	0.66	0.79
DL-Methionine	0	0.09	0.21	0.23	0	0.09	0.21	0.23
L-Histidine	0.23	0.27	0.25	0.28	0.23	0.27	0.25	0.28
Vitamin premix	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix	0.15	0.15	0.15	0.15	0.15	0.15	0.15	0.15
Choline chloride	0.20	0.20	0.20	0.20	0.20	0.20	0.20	0.20

Table2, Composition of aquafeeds for rainbow trout, detailing the ingredients and their respective inclusion levels (expressed as a percentage of the total feed) for both control and test diets.

Rainbow trouttrials	Control			Test		
Pellet size	3 & 4.5 mm	6 & 9 mm	9 mm	3 & 4.5 mm	6 & 9 mm	9 mm
protein-to-fat ratios	44/28	42/30	38/32	44/28	42/30	38/32
Pea protein concentrate	2.51	4.98	6	2.51	4.98	6
Soy protein concentrate	8	8	8	8	8	8
Fishmeal LT	19.64	14.62	12.50	19.64	14.62	12.50
Fishmeal SA	10	10	10	10	10	10
Poultry meal	8	8	8	3	3	3
Hydrolysed poultry meal	0	0	0	5	5	5
Hydrolysed feather meal	6	6	6	6	6	6
Rapeseed oil	14.66	16.95	18.45	14.66	16.95	18.45
Fish oil	8	8	9	8	8	9
Emulsifier	0.30	0.40	0.40	0.30	0.40	0.40
Wheat	11	11	14.73	11	11	14.73
Corn gluten	8	8	2.18	8	8	2.18
Pea starch	1	1	1	1	1	1
Diamol	1	1	1	1	1	1
Monoammonium phosphate	1.04	0.66	0.89	1.04	0.66	0.89
L-Lysine	0.12	0.53	0.79	0.12	0.53	0.79
DL-Methionine		0.08	0.20		0.08	0.20
L-Histidine	0.13	0.18	0.26	0.13	0.18	0.26
Vitamin premix	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix	0.15	0.15	0.15	0.15	0.15	0.15
Choline chloride	0.20	0.20	0.20	0.20	0.20	0.20

Table3, Chemical composition of aquafeeds for both rainbow trout and Atlantic salmon. It includes general analysis (Table A), amino acid analysis (Table B), and fatty acid analysis (Table C).

Diet	Atlantic salmon		Rainbow trout	
	Test	Control	Test	Control
Ash (%)	7,9	8,0	7,6	8,2
Kjeldahl-N (%)	6,1	6,0	6,1	6,1
Total Fat (%)	30,0	*	29,5	30,8
Protein	38,1	37,6	38,1	37,8
Energy (MJ/kg)	23,6	23,9	23,9	23,9
Astaxanthin (mg/kg)	26,0	26,0	45,0	42,0

A

Amino Acids (g/kg)	Atlantic salmon		Rainbow trout	
	Test	Control	Test	Control
Cysteine	3,9	4,0	3,4	3,5
Methionine	7,4	7,5	7,8	5,7
Aspartic Acid	28,5	27,5	29,4	29,0
Threonine	12,9	13,2	13,0	12,7
Serine	16,0	16,2	14,5	15,5
Glutamic acid	50,0	48,2	58,5	49,6
Proline	17,2	17,0	17,5	16,6
Glycine	17,2	18,1	15,8	17,4
Alanine	16,3	16,5	17,2	16,4
Valine	15,8	15,9	15,4	15,6
Isoleucine	14,0	14,0	14,3	14,1
Leucine	23,8	23,9	26,5	23,5
Tyrosine	8,0	8,0	8,5	7,7
Phenylalanine	13,3	12,7	14,1	12,9
Histidine	8,4	9,1	9,5	8,7
Lysine	26,5	25,5	25,6	26,2
Arginine	22,4	23,3	23,1	22,5
Tryptophan	3,2	3,1	3,0	3,1

B

Fatty Acids (g/kg)	Atlantic salmon		Rainbow trout	
	Test	Control	Test	Control
C12:0	0,3	0,2	0,1	0,2
C14:0	3,9	4,7	3,2	3,7
C14:1n7	0,1	0,1	0,03	0,1
C15:0	0,4	0,5	0,3	0,4
C16:0	25,0	25,9	23,1	26,0
C16:1n7	4,5	5,4	3,5	4,2
C17:0	0,4	0,4	0,3	0,4
C18:0	9,2	9,4	8,9	10,1
C18:1n9t	0,3	0,3	0,2	0,3
18:1n9c	131,3	117,4	132,0	138,1
C18:2n6c	45,0	40,6	47,0	47,3
C20:0	1,7	1,5	1,6	1,7
C18:3n6	0,3	0,3	0,2	0,3
C20:1	7,0	7,9	5,7	6,4
C18:3n3	16,4	14,2	17,7	17,9
C20:2	2,1	2,7	1,7	2,0
C22:0	1,8	1,5	1,6	1,7
C20:3n6	0,2	0,2	0,1	0,2
C22:1n11	6,2	7,9	4,5	5,3
C22:1n9	1,1	1,0	0,8	1,1
C20:3n3	0,3	0,4	0,3	0,5
C20:4n6	0,8	0,9	0,8	0,9
C22:2	0,6	0,7	0,5	0,6
C24:0	0,3	0,3	0,3	0,4
C20:5n3	5,9	6,0	5,9	6,6
C24:1	0,8	0,9	0,8	0,9
C22:5n3	1,0	1,0	0,9	1,0
C22:6n3	9,2	8,2	9,8	10,9

C

17

3.3 Sampling procedures in fish lab

The process of assessing visceral fat involved scoring its visibility on a scale from 1 to 5, with 1 indicating clearly visible fat and 5 meaning the fat was not visible at all (Fig. 4). Additionally, fat deposition on the surface of each fish's heart was examined and scored on a scale from 0 to 3 (Fig. 5).
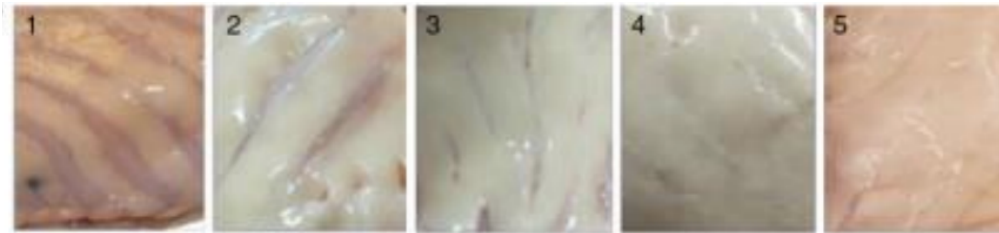

Fig4. Scale scoring for visual determination of fat accumulation on pyloric caeca in Atlantic salmon(Mørkøre et al., 2020).
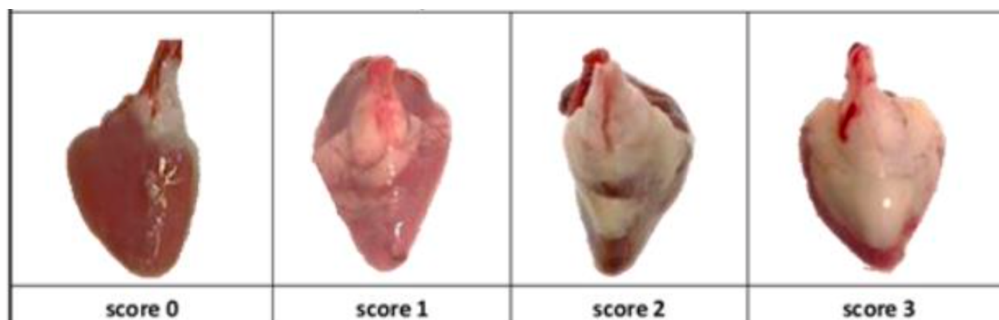

Fig5. Scale scoring for visual determination of fat accumulation on the cardiac surface in Atlantic salmon(Formanowicz, 2022).

3.6 Fillet quality analyses

3.6.1 Gaping

The assessment of fillet gaping involved measuring both the number and size of slits within each fillet as outlined in table 4. The evaluation method included gently placing a flat palm beneath the fillet to identify any splits or holes post-filleting. The severity of gaping was rated on a scale from 0 to 5, as described by Andersen (Andersen et al., 1994). On this scale, a score of 0 indicated no visible slits or holes. A score of 1 was assigned for fillets with fewer than five small slits (less than 2 cm), and a score of 2 for those with fewer than ten small slits. Fillets exhibiting more than ten small slits or some larger slits (more than 2 cm) received a score of 3. Fillets with significant gaping or those that were falling apart were rated either 4 or 5 for extreme cases of gaping, as detailed in Table 4.

Table4, Classification scale for fillet gaping(Andersen et al., 1994)

Score	Description
0	No gaping
1	Few small slits < 5
2	Some small slits < 10
3	Many slits >10 or a few large slits(>2cm)
4	Severe gaping (Many large slits)
5	Extreme gaping (the fillet falls apart)

Small slits < 2 cm
Large slits > 2 cm

3.6.2 Dark stained segments

In this study, the number of dark stained segments were counted on each fillets. The focus was on identifying and outlining areas of dark stained segments on the surface of Atlantic salmon and rainbow trout fillets.

3.6.3 Chemical analyses

In this study, each fish fillet sample was processed using the Retsch GM200 to achieve a consistent texture by homogenizing at 5000 RPM for ten seconds (fig.6). This precise and uniform preparation method was critical for ensuring accurate chemical analysis. The samples were then analyzed in detail to measure fat content and astaxanthin levels, and to assess the fatty acid profiles of fillets.

Fig6. Homogenization of fish fillet with Retsch GM200 in Labtek,NMBU

For homogenization in this study, the NQC part was selected for grinding. This area, precisely highlighted and marked, followed a standardized processing protocol to ensure uniform sample preparation for chemical analyses.
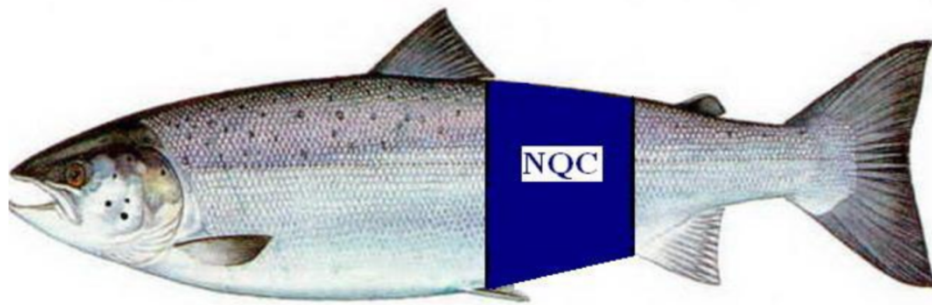

Fig7. Sampling area for analysis of fat and astaxanthin (Rahnama, 2010).

3.6.4 Fat content analysis

The fat content analysis was carried out at LabTek, an expert laboratory in the fields of livestock and aquaculture at NMBU, following the protocols stipulated by Commission Regulation (EC) No 152/2009. The procedure was executed using the state-of-the-art Soxtec™ 8000 system, which automates the fat extraction process using petroleum ether by implementing the Soxhlet extraction technique (fig.8). The sample preparation involved enclosing the specimens in cellulose thimbles measuring 33x80 mm and submerging them in a solvent bath maintained at temperatures ranging from 40 to 60 degrees Celsius. Following the extraction phase, the resulting fat extract was carefully transferred into aluminum containers and subjected to a drying process in an oven set at 103 degrees Celsius for 30 minutes, ensuring complete solvent evaporation. The final step involved accurately quantifying the residual fat through gravimetric analysis, thereby providing a precise measure of fat content. This methodical approach ensures the reliability and reproducibility of the fat content data, crucial for understanding the nutritional quality of aquaculture products.

Fig8. The fat content analysis via art Soxtec™ 8000 at LabTek

3.6.5 Astaxanthin analysis

The astaxanthin analysis was conducted at LabTek, specifically the Analysis Lab for Livestock and Aquaculture, at NMBU. The method employed was the "CEN/TS 16233-1:2011 (E) - HPLC method for the determination of xanthophylls in fish flesh. Part 1: Determination of astaxanthin and canthaxanthin." The procedure commenced by weighing approximately 1.5 mg of homogenized fish flesh and 1 g of BHT (2,6-Di-tert-butyl-p-cresol) into a volumetric flask. Subsequently, 5 mL of tetrahydrofuran was added, and the flask was filled to the mark with tetrahydrofuran. A portion of 10 mL from this solution was then transferred to a 100 mL volumetric flask, followed by the addition of 85 mL of heptane. The analysis utilized an Ultimate 3000 UHPLC system with a UV detector (Thermo Scientific) to measure the concentration of astaxanthin.

3.6.6 Fatty acid composition (FAME) analysis

The fatty acid analysis was conducted at the Faculty of Biosciences, NMBU, utilizing the method outlined in the document "Msp 1046 Fatty acid composition" developed by BIOVIT. This method involves synthesizing fatty acids into fatty acid methyl esters (FAME) and extracting them directly from various fresh organic materials such as tissues, oils, and feeds. Notably, this process does not require an initial organic solvent extraction, making it simpler and more time-efficient. The innovative aspect of this method allows for the inclusion of up to 33% water in the sample material during the FAME synthesis, accommodating even wet samples.The analysis utilized the Trace GC Ultra with an auto injector (Thermo Scientific) as the main instrument. This process is based on a direct method for fatty acid methyl ester synthesis, significantly modified from the original technique described by O'Fallon, J.V. in 2007, where the volumes have been scaled down for efficiency (O'Fallon et al., 2007).

3.6.7 Myocommata width measurement by Image J Analysis

Attention to the belly flap region, acknowledged for its high adipocyte concentration, permits the prediction of lipid levels through digital image analysis. This method of analysis, characterized by Einen et al. (1998), provides an estimation of fat content within this region. The procurement of digital imagery of these sections is enabled by the utilization of advanced imaging devices, including Photo light boxes and digital cameras (Einen et al., 1998).

After image acquisition, analytical tasks are undertaken with software such as ImageJ, which facilitates the quantification of lipid content. Variability in lipid content, which manifests seasonally and across different individuals and species, is attributed to a range of environmental conditions and dietary regimens (Alanärä et al., 2001).The analysis for the width of myocommata in the left-side fillets is conducted using ImageJ software, enabling precise morphometric assessments.

A standardized region for measurement was established to ensure consistency across samples. This involved drawing a line from the start of the dorsal fin, perpendicular to the lateral line, and marking a point 15 mm below it. Within this area, a rectangle measuring 15 mm by 45 mm was defined as the zone for analysis (fig.9).

In this predefined zone, 20 points were randomly selected to measure the myocommata width, aiming to minimize bias. The average width from these points was calculated to represent the

myocommata width of each fillet. This streamlined approach ensures accurate and consistent measurements across all samples.
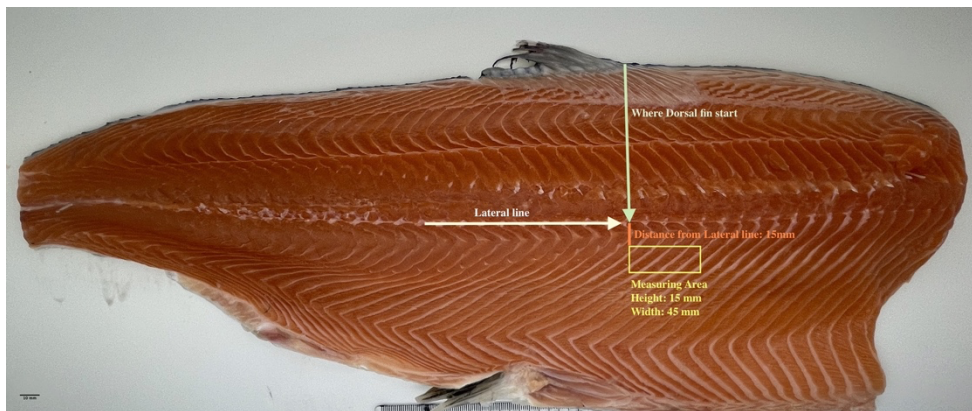

Fig9, The image shows a method for determining a measuring area on a fish fillet. A lateral line on the fillet is identified and labeled. Starting from this line, a distance of 15 mm is measured outward. At this point, a rectangular area for measurement is established, with dimensions labeled as 45 mm in width and 15 mm in height. This process outlines the specific region on the fillet to be analyzed or used for further testing.

3.8 Statistical analyses

Statistical analyses were carried out using SAS software (version 9.5, SAS Institute, Cary, NC, USA). Significant differences between dietary treatments and species were determined using pdiff and Duncan's multiple range test. We accounted for the potential effects of gender, body weight imbalances, and farming location. The significance level was set at 5% ($P \leq 0.05$).

3.9 Calculations

The biometric characteristics of Atlantic salmon and rainbow trout were carefully evaluated during both sampling occasions. Detailed measurements of their body weight, fork length, gutted weight, and fillet weight were meticulously recorded. A platform scale was used to determine these weights and lengths. Using this data, specific formulas were applied to calculate the slaughter yield, condition factor, cardiomatic index, hepatosomatic index, and fillet yield.

Slaughter yield (%) was calculated as:

- Slaughterd Yield (%) = (Gutted weight (g) × 100) / Body weight (g)

Condition Factor (CF) was calculated as:

- Condition Factor = (Round body weight (g) × 100) / (Body length)^3

Fillet Yield (FY) was calculated as:

- Fillet Yield (%) = (Fillet weight (g) × 100) / Body weight (g)

Cardiosomatic Index (CSI) was calculated as:

- Cardiosomatic Index (CSI) = (Heart weight (g)) × 100) / Body weight (g)

Hepatosomatic Index (HSI) was calculated as:

- Hepatosomatic Index(HSI) = (Liver weight (g)) × 100) / Body weight (g)

4. Results

4.1 Biometric traits

Biometric traits for rainbow trout and Atlantic salmon fed with control and test diets were assessed (table 5). For rainbow trout, those on the test diet had a significantly higher (P=0.001) average body weight of 2120±159 g compared to 1893±96 g in the control group at the first sampling (March 2023). Similarly, gutted weight was higher in the test group at 1808±138 g, compared to 1643±83 g in the control group (P=0.001). However, there were no significant differences in fillet weight between the test group 1349 ± 113 g, and the control group1397 ± 68 g, with a p-value of 0.3. For Atlantic salmon, no significant differences in body weight were observed between the test 1420±87 g and control groups 1436±82 g at the first sampling time (June 2023). In the second sampling (December 2023), significant differences were noted in body weight for rainbow trout, with the control group weighing 3725 ± 170 g compared to 3446 ± 204 g for the test group (P=0.01). However, no other biometric traits showed significant differences. For Atlantic salmon, no significant differences in biometric traits were found in either sampling.

The study also compared the two species. In the first sampling, rainbow trout showed significantly higher body and gutted weights than Atlantic salmon (P < 0.001). Additionally, the fillet weight of rainbow trout was significantly higher than that of Atlantic salmon (P < 0.001). By the second sampling, although initial differences in body and gutted weight between the species had diminished, a significant difference in fork length emerged, with Atlantic salmon exhibiting longer forks than rainbow trout (P < 0.01).

Table5. Whole body weight, gutted weight, fillet weight, and fork length of Atlantic salmon and rainbow trout fed control or test feed. Values are expressed as means ± standard errors (SE). Different superscripts indicate significant differences between the dietry groups (P≤0.05).

Biometric traits	First Sampling				P-value
	Atlantic Salmon		Rainbow trout		
	Control	Test	Control	Test	
Body weight (g)	1436±82.3[b]	1421±87.4[b]	1894±96[a]	2120±159.4[a]	<0.001
Gutted weight (g)	1258±82.4[b]	1244±86.5[b]	1644±83.9[a]	1808±138.8[a]	<0.001
Fork length (cm)	47.6±0.7	47.6±0.8	47.4±0.8	48±1.3	0.94
Fillet weight (g)	1397±68	1349±113	917±66	915±67	<0.001
Second Sampling					
Body weight (g)	3806±103.7	3829±101.9	3726±170	3465±204.7	0.11
Gutted weight (g)	3380±93.8	3367±117.1	3120±151.4	3094±204.3	0.06
Fork length (cm)	66.3±0.6[a]	66.6±0.6[a]	55±0.8[b]	54.5±0.9[b]	<0.001
Fillet weight (g)	2331±11	2326±164	2353±80	2319±90	0.16

4.2 Chemical analyses
4.2.1 Fat Content Analysis

The fat content was assessed in fillets of rainbow trout and Atlantic salmon at two different sampling times (table.6). at the first sampling, rainbow trout showed a fat content of 13 ± 1.03 while Atlantic salmon recorded of 10.4 ± 0.73; the difference in values yielded a non-significant p-value of 0.06. In the second sampling, rainbow trout demonstrated a fat content of 14.6 ± 0.52 compared to 13.04 ± 0.52 for Atlantic salmon, with this difference also proving to be statistically non-significant with a p-value of 0.1.

No significant differences were observed in rainbow trout or Atlantic salmon fed with control and test diets (table.7).

Table 6.Mean total fat content in fillets of rainbow trout and Atlantic salmon during the first and second sampling. Values are expressed as means ± standard errors (SE). The p-values indicate the statistical significance of differences between the species for each sampling.

Total fat	Rainbow trout	Atlantic salmon	p value
First sampling	13±1.03	10.4±0.73	0.06
Second sampling	14.6±0.52	13.4±0.52	0.1

Table 7.Mean total fat content in fillets of rainbow trout and Atlantic salmon fed control or test feed. Values are expressed as means ± standard errors (SE). The p-values indicate the statistical significance of differences between the species for each sampling.

Total fat	Rainbow trout			Atlantic salmon		
	Control	Test	p value	Control	Test	p value
First sampling	13.1±1.17	13.1±1.17	1	10.6±1.25	10.3±1.25	0.8
Second sampling	14.4±1.1	14.8±1.1	0.8	12.9±0.38	13.9±0.38	0.2

4.2.2 Astaxanthin

The astaxanthin content was assessed in rainbow trout and Atlantic salmon at two different sampling points (table.8). During the first sampling, rainbow trout exhibited a mean astaxanthin level of 10.4 ± 0.21 mg/kg, being significantly higher than Atlantic salmon, which recorded a mean of 3.2 ± 0.16 mg/kg ($p < 0.001$). In the second sampling, rainbow trout displayed a mean astaxanthin level of 12.2 ± 0.41 mg/kg, while Atlantic salmon showed a mean of 6 ± 0.41 mg/kg. Rainbow trout consistently demonstrated higher astaxanthin levels compared to Atlantic salmon across both samplings, with a statistically significant difference observed at both sampling points($p < 0.001$). No significant differences were observed in rainbow trout or Atlantic salmon fed with control and test diets (table.9).

Table 8. Mean Axtaxanthin for rainbow trout and Atlantic salmon at first and second sampling points. Values are expressed as means ± standard errors (SE). The p-values indicate the statistical significance of differences between the species for each sampling.
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Astaxanthin	Rainbow trout	Atlantic salmon	p value
First sampling	10.4±021	3.2±0.16	<0.001
Second sampling	12.2±0.41	6±0.41	<0.001

Table 9. Mean Axtaxanthin for rainbow trout and Atlantic salmon fed control or test feed.Values are expressed as means ± standard errors (SE). The p-values indicate the statistical significance of differences between the species for each sampling.

Astaxanthin	Rainbow trout			Atlantic salmon		
	Control	Test	p value	Control	Test	p value
First sampling	10.3±0.32	10.6±0.32	0.6	3.05±0.22	3.42±0.22	0.3
Second sampling	12.2±0.68	12.1±0.68	0.8	5.6±0.64	6.4±0.64	0.4

4.2.3 Fillet Fatty Acid Composition

The fatty acid composition in Atlantic salmon and rainbow trout muscle was carried out to understand the distinct responses within and between species to dietary changes (table8, table9). In Atlantic salmon, alterations in diet between the test and control groups generally did not result in statistically significant changes in fatty acid levels. However, there was a significant decrease in C22:1n11 levels in the test diet (p = 0.03) and significantly higher levels of C20:2 in the test group.

Conversely, significant differences in specific fatty acids were observed in rainbow trout, including increased levels of C14:0 (p = 0.04) and decreased levels of C16:1n7 (p = 0.01) in the test diet. Comparative analysis between the species highlighted significant differences in most fatty acids, Notably, rainbow trout exhibited significantly higher levels of C14:0, C16:0, C16:1n7, C18:1n9, C18:2n6c, C22:2, C20:5n3, and C22:6n3 (p < 0.001). In contrast, Atlantic salmon had significantly higher levels of C20:1 and C20:2 than rainbow trout (p < 0.001).

Table 8. Displays the concentrations of various fatty acids (g/kg) in Atlantic salmon and rainbow trout fed test and control diet. Values are presented as mean ± standard deviation. P-values are provided to indicate the statistical significance of the differences between the test and control groups for each fatty acid within the species.

Fatty Acids(g/kg)	Atlantic salmon			Rainbow trout		
	Test	Control	p value	Test	Control	p value
C14:0	1.63±0.02	1.74±0.04	0,13	2.25±0.05	2.80±0.10	0,04
C16:0	11.2±0.31	10.95±0.16	0,85	14.35±0.65	17.25±0.45	0,07
C16:1n7	1.75±0.04	1.94±0.05	0,10	3.40±0.10	4.60±0.10	0,01
C18:0	3.54±0.05	3.30±0.05	0,07	3,90	4.60±0.20	0,07
C18:1n9t	0.21±0.04	0.17±0.04	0,47	0.25±0.05	0.25±0.05	1
C18:1n9	61.25±2.16	56.54±1.57	0,22	59.15±2.35	68.35±1.25	0,07
C18:2n6c	19.59±0.69	18.09±0.56	0,23	17.70±1	20.25±0.25	0,13
C20:1	5.35±0.23	4.65±0.10	0,11	4.35±0.05	5.45±0.25	0,05
C18:3n3	6.38±0.29	6.16±0.19	0,60	6.05±0.35	6.85±0.05	0,15
C20:2	2.51±0.08	2.05±0.07	0,05	1.75±0.05	2.0±0.10	0,15
C22:1n11	2.11±0.03	2.42±0.07	0,03	2.65±0.05	3.70±0.30	0,07
C22:2	0	0	0	1.02±0.01	1.20±0.09	0,19
C20:5n3	1.97±0.01	2.04±0.05	0,34	2.50±0.10	2.95±0.15	0,13
C22:6n3	5.69±0.11	5.47±0.17	0,38	6.60±0.40	7.25±0.25	0,30
Sum n1	71.37±2.30	66.57±1.67	0,21	70.77±2.29	83.25±2.08	0,06
Sum n3	16.02±0.51	15.22±0.42	0,35	16.73±0.74	18.87±0.53	0,14
Sum n6	20.27±0.60	18.60±0.57	0,18	18.56±0.83	20.97±0.29	0,11
Sum n0	17.79±0.03	17.22±0.28	0,19	22.32±0.11	26.01±0.87	0,05
EPA+DHA	7.66±0.12	7.52±0.22	0,62	9.06±0.47	10.24±0.38	0,19

Table 9. This table summarizes the fatty acid content (g/kg) on fillet for Atlantic salmon and rainbow trout. The data is presented as mean ± standard deviation for various fatty acids. The table also includes p-values to highlight statistically significant differences between the species for each fatty acid listed.

Fatty acid (g/kg)	Species		p value
	Atlantic salmon	Rainbow trout	
C14:0	1.68±0.03	2.52±0.17	<0.001
C16:0	10.98±0.14	15.8±0.90	<0.001
C16:1n7	1.84±0.06	4.0±0.35	<0.001
C18:0	3.41±0.08	4.25±0.22	0,06
C18:1n9t	0.19±0.02	0.25±0.03	0,28
C18:1n9	58.89±1.74	63.75±2.87	<0.001
C18:2n6c	18.84±0.56	18.97±0.85	0,01
C20:1	5.0±0.23	4.9±0.33	<0.001
C18:3n3	6.27±0.15	6.45±0.27	0,08
C20:2	2.27±0.14	1.87±0.09	<0.001
C22:1n11	2.26±0.09	3.17±0.33	0,08
C22:2	0	1.11±0.06	<0.001
C20:5n3	2.0±0.03	2.72±0.15	0,01
C22:6n3	5.58±0.10	6.92±0.27	0,04
Sum n1	69.15±1.89	77.0±3.82	<0.001
Sum n3	15.61±0.36	17.8±0.72	0,46
Sum n6	19.43±0.59	19.76±0.78	<0.001
Sum n0	17.50±0.20	24.16±1.12	<0.001
EPA+DHA	7.58±0.11	9.65±0.42	0,01

4.2.4 Heart Fatty Acid Composition

No significant differences were observed between the test and control groups for any fatty acids in both Atlantic salmon and rainbow trout. Nonetheless, a significant distinction was found in the levels of Stearic acid (C18:0); Atlantic salmon in the test group exhibited a Stearic acid content of 2.1 ± 0.17 g/kg, significantly higher than the 1.4 ± 0.17 g/kg recorded for rainbow trout in their respective test group (P=0.05) as indicated in Table 10. Additionally, the concentration of Arachidonic acid (C20:4n6) showed a significant variation between the species across both control and test groups. Specifically, the test group of Atlantic salmon had a higher level of C20:4n6 (0.48 ± 0.02 g/kg) compared to the test group of rainbow trout (0.32 ± 0.02 g/kg), with the difference being statistically significant (P < 0.01). In the control groups, the Atlantic salmon also registered a significantly higher concentration of C20:4n6 (0.46 ± 0.02 g/kg) compared to the rainbow trout (0.26 ± 0.02 g/kg) (P = 0.002).

Regarding the total fatty acid content in the heart tissues of the control groups, Atlantic salmon had a higher fatty acid content at 37.2 ± 3.86 compared to 23.7 ± 3.86 in rainbow trout. Although this difference suggests higher fatty acid levels in Atlantic salmon, it did not reach statistical significance (P=0.07).

Table 10. This table summarizes the fatty acid content (g/kg) on heart for Atlantic salmon and rainbow trout. The data is presented as mean ± standard deviation for various fatty acids. The table also includes p-values to highlight statistically significant differences between the species for each fatty acid listed.

Fatty Acid (g/kg)	Atlantic salmon		rainbow trout		P value between species
	Control (n=32)	Test (n=32)	Control (n=16)	Test (n=16)	
C14:0	0.4±0.13	0.6±0.13	0.4±0.13	0.4±0.13	0,28
C16:0	5.3±0.45	5.8±0.45	3.9±0.45	4.3±0.45	0,01
C16:1n7	0.4±0.08	0.5±0.08	0.4±0.08	0.4±0.08	*
C17:0	0.2±0.16	0.3±0.16	0.04±0.16	0.04±0.16	*
C18:0	1.8±0.17	2.1^a±0.17	1.2±0.17	1.4^b±0.17	0,01
18:1n9	12.8±1.87	15.1±1.87	6.7±1.87	8.9±1.87	0,01
C18:2n6	4.2±0.65	3.8±0.65	2.3±0.65	3.1±0.65	0,09
C18:3n3	1.5±0.24	1.3±0.24	0.8±0.24	1.3±0.24	0,26
C20:2	0.5±0.07	0.5±0.07	0.2±0.07	0.3±0.07	0,03
C20:3n6	0.2±0.03	0.2±0.03	0.1±0.03	0.1±0.03	0,06
C22:1n11	0.5±0.1	0.4±0.1	0.3±0.1	0.4±0.1	0,31
C20:3n3	0.2±0.07	0.3±0.07	0.1±0.07	0.1±0.07	0,07
C20:4n6	0.5^a±0.02	0.5^a±0.02	0.3^b±0.02	0.3^b±0.02	<0.001
C22:6n3	5.3±0.64	4.5±0.64	5 ±0.64	5.5±0.64	0,6
Σn-6 PUFA	5.8±0.63	5.8±0.63	2.9±0.63	3.9±0.63	0,05
Σn-3 PUFA	8.6±0.9	7.7±0.9	7.1±0.9	8.2±0.9	0,59
ΣMUFA	15.1±2.28	18.4±2.28	8.1±2.28	10.6±2.28	0,04
ΣSFA	7.9±1.18	10.6±1.18	5.6±1.18	6.3±1.18	0,02
EPA+DHA	6.5±0.72	5.7±0.72	5.8±0.72	6.4±0.72	0,95

4.3 Myocommata width

The myocommata width in Atlantic salmon and rainbow trout on different diets was measured at two sampling points (Fig.12). Initially, rainbow trout on the test diet had a myocommata width of 0.98 ± 0.03 mm, compared to 1.01 ± 0.06 mm in the control group, with no significant difference ($p = 0.77$). Similarly, Atlantic salmon showed no difference in myocommata width between the test and control diets, both averaging 0.88 ± 0.05 mm ($p = 0.93$).

In the second sampling, Atlantic salmon on the test diet had a width of 1.55 ± 0.07 mm, compared to 1.42 ± 0.09 mm for the control diet, again with no significant difference ($p = 0.25$). Rainbow trout had widths of 1.92 ± 0.08 mm for the test diet and 1.76 ± 0.07 mm for the control diet, also not significantly different ($p = 0.33$).

Comparing the species, there were significant differences in myocommata widths between rainbow trout and Atlantic salmon. In the first sampling, a P-value of 0.03 indicated a significant difference, which was even more pronounced in the second sampling with a P-value of 0.001.
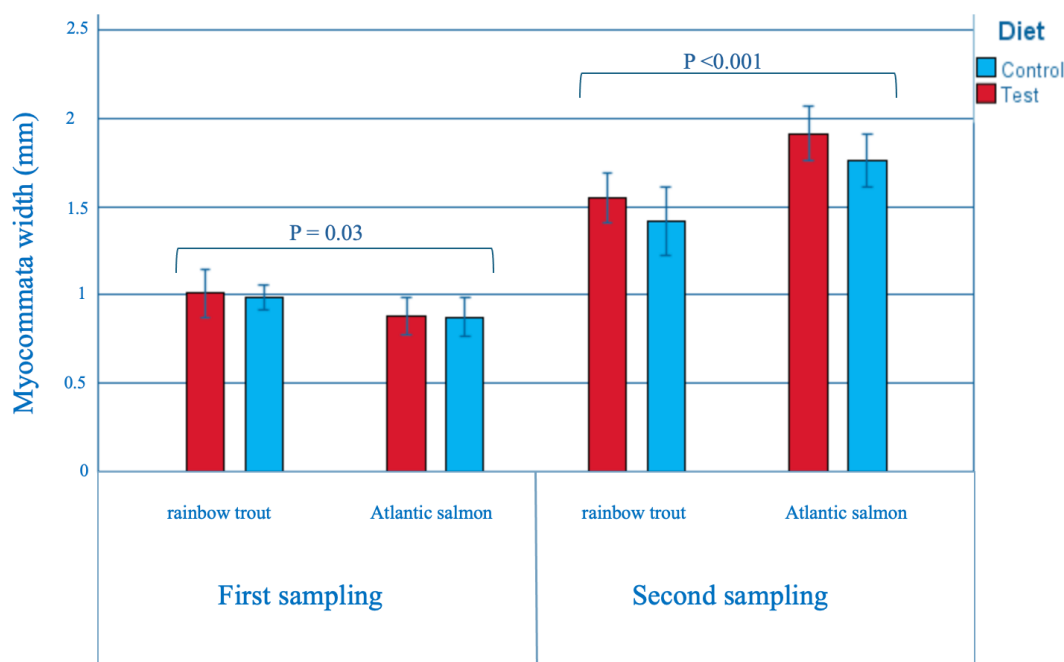

Fig12, Mean myocommata width measurements of Atlantic salmon and rainbow trout fed control or test feed, assessed at two sampling points. The data are represented as mean values with their respective standard errors (SE). The table also includes p-values to highlight statistically significant differences between groups at a threshold of $p \leq 0.05$.

4.4 Correlation between myocommata and fork length

The scatter plot illustrate the relationship between fork length and myocommata width for rainbow trout and Atlantic salmon, depicted with blue and orange dots, respectively (fig.13). For rainbow trout, the regression equation was $y=0.0416x-0.887$, with a coefficient of determination 43.3%. This demonstrate a positive correlation, indicating that as the fork length increased, the myocommata width also tended to increase ($P < 0.01$).

similarly, Atlantic salmon showed a positive correlation between these two variables, as indicated by the regression equation $y=0.0488x-1.424$ and a R^2 value of 0.72. This higher R^2 value implied that 72.49% of the variability in myocommata width was attributable to variations in fork length ($P < 0.01$).

Fig13. Comparative analysis of myocommata width in relation to fork length in rainbow trout and Atlantic salmon fed control or test feed, assessed at two sampling points.
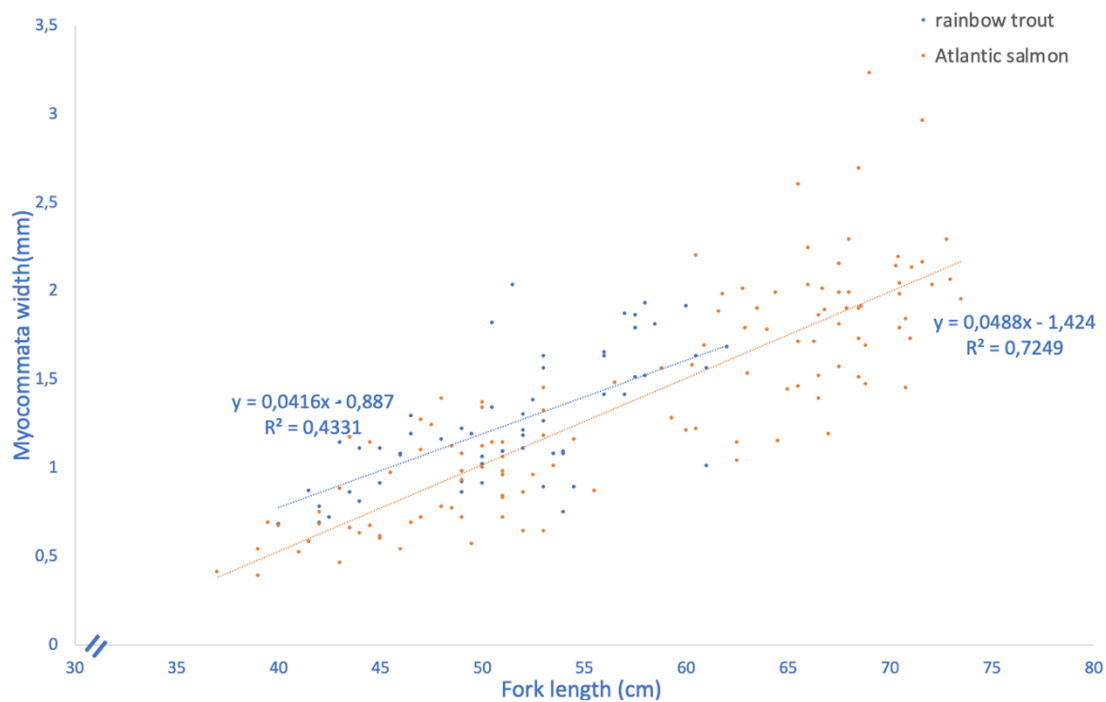
4.5 Hepatosomatic Index

The hepatosomatic index (HSI), was measured in Atlantic salmon and rainbow trout subject to different diets. This index was assessed at two separate time points to determine the influence of diet over time (fig.14). For the rainbow trout, the HSI in the first sampling exhibited a range of $1.19 \pm 0.05\%$ in the test group compared to $1.29 \pm 0.04\%$ in the control group, a difference that was not statistically significant ($p = 0.1$). In the second sampling, the HSI of rainbow trout ranged from $1.30 \pm 0.04\%$ in the test group to $1.43 \pm 0.06\%$ in the control group, also without a significant difference ($p = 0.2$). For Atlantic salmon, the first sampling revealed an HSI of $1.12 \pm 0.03\%$ in the control group and $1.13 \pm 0.03\%$ in the test group, indicating no significant difference ($p = 0.7$). At the second sampling, the HSI values for Atlantic salmon were $1.76 \pm 0.06\%$ for the control diet group and $1.61 \pm 0.05\%$ for the test diet group, again showing no significant difference ($p = 0.1$). Apart from evaluating the effects of control and test diets, an assessment between species was also conducted. During the first sampling, there was a statistically significant difference in the HSI between rainbow trout and Atlantic salmon, as indicated by a P-value of 0.003. By the second sampling, the differences between the species in terms of HSI showed P-value of less than 0.001.
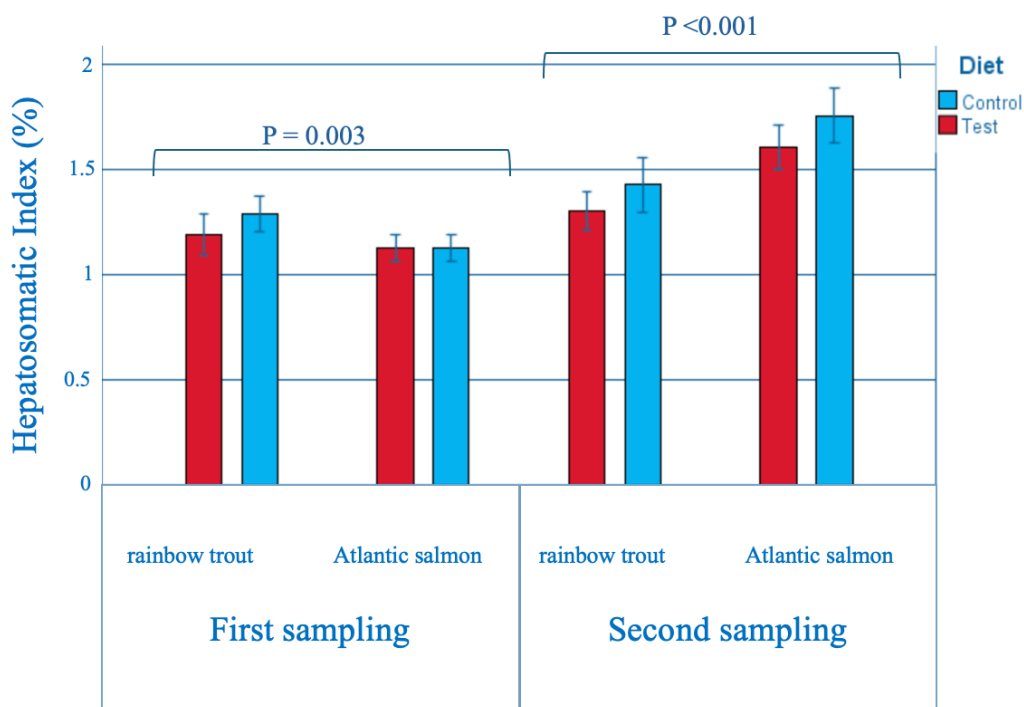

Fig14. Hepatosomatic index (HSI) % ,of Atlantic salmon and rainbow trout, fed control or test feed, assessed at two sampling points. The data are represented as mean values with their respective standard errors (SE). The table also includes p-values to highlight statistically significant differences between groups at a threshold of p ≤ 0.05.

4.6 Cardiosomatic Index

The cardiosomatic index (CSI), was assessed in Atlantic salmon and rainbow trout fed with control and test diets (fig.15). In the first sampling for rainbow trout, the CSI did not differ significantly between the test and control groups, both demonstrating a value of $0.09 \pm 0.01\%$. The second sampling also showed no significant differences, with a CSI of $0.08 \pm 0.001\%$ for both diets ($p = 0.836$).

For Atlantic salmon, initial samples showed a consistent CSI of $0.11 \pm 0.001\%$ across diets, with no significant difference ($p = 0.882$). However, later measurements indicated a significant increase in CSI to $0.12 \pm 0.001\%$ in the test diet group, compared to a stable $0.11 \pm 0.001\%$ in the control group ($p = 0.029$).

Apart from diets, a comparative assessment between species was also conducted. In both the first and second sampling, the Cardiomatic Index (CSI) was significantly higher in Atlantic salmon compared to rainbow trout, with a P-value of less than 0.001 in each instance.
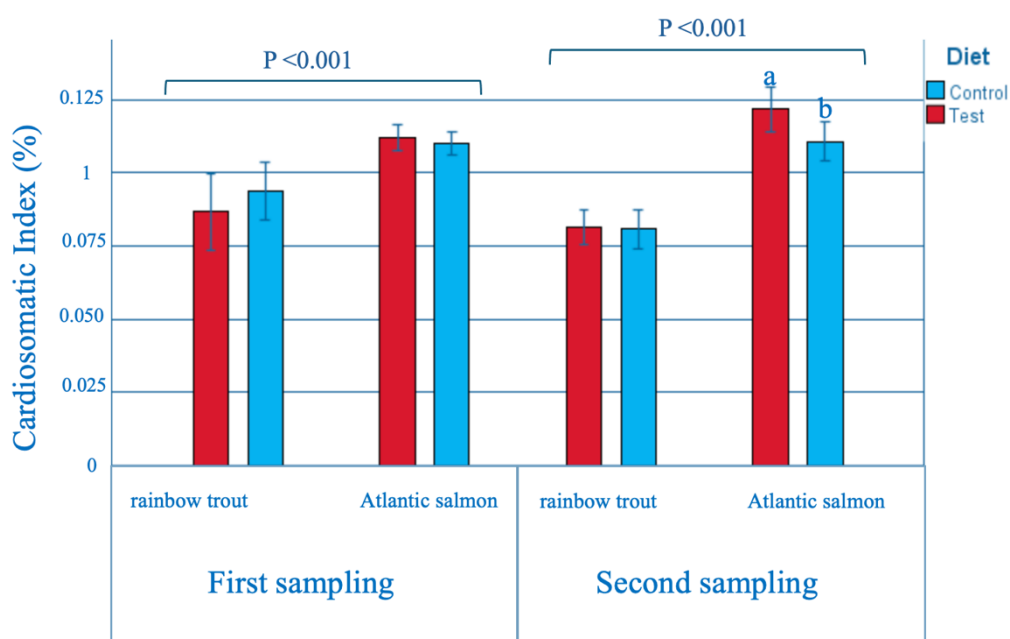

Fig15.Cardiosomatic index (CSI)%, of Atlantic salmon and rainbow trout, fed control or test feed, assessed at two sampling points.The data are represented as mean values with their respective standard errors (SE). Distinct superscripts denote statistically significant differences between groups at a threshold of $p \leq 0.05$.

4.7 Conditional Factor

The Conditional Factor (CF) was evaluated in Atlantic salmon and rainbow trout fed with control and test diets in two sampling time (fig.16). For rainbow trout, initial measurements of the CF presented a mean value of 1.89 ± 0.09 within the test group, compared to 1.78 ± 0.07 in the control group, showed a nonsignificant statistical difference (p = 0.763). In the second sampling, CF ranged from 2.00 ± 0.07 in the test group to 2.28 ± 0.06 in the control group, marking a statistically significant difference (p = 0.009). Conversely, the assessment of Atlantic salmon during the first sampling indicated a CF of 1.30 ± 0.08 for the control group and 1.23 ± 0.04 for the test group, marking a statistically non significant difference (p = 0.942). In the second sampling CF values at $1.31 \pm 0.02\%$ for the control group and 1.28 ± 0.02 for the test group; displayed any significant difference (p = 0.316).

Apart from diets, an assessment between species was also conducted. In the first sampling, there was a statistically significant difference in the CF between rainbow trout and Atlantic salmon, with a P-value of less than 0.001. Similarly In the second sampling, there was a statistically significant difference in the CF between rainbow trout and Atlantic salmon, with a P-value of less than 0.001.
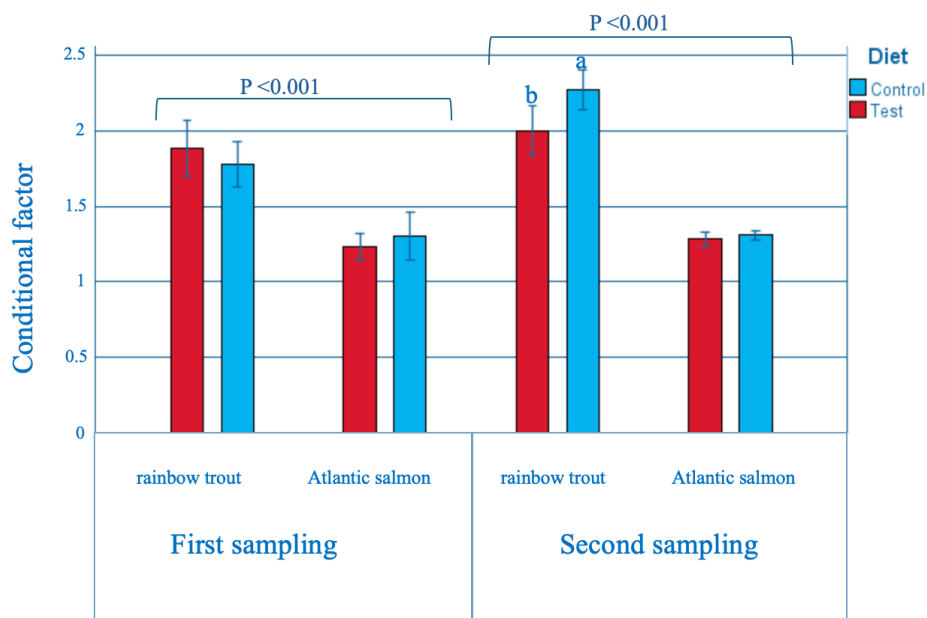

Fig16,Conditional Factor (CF) of Atlantic salmon and rainbow trout, fed control or test feed, assessed at two sampling points. The data are represented as mean values with their respective standard errors (SE). Distinct superscripts denote statistically significant differences between groups at a threshold of $p \leq 0.05$.

4.7 Slaughter Yield (%)

The slaughter yield (SY) was assessed for both Atlantic salmon and rainbow trout on test and control diets at two time points. As illustrated in Figure 17, the first sampling for rainbow trout showed SY was $85.2 \pm 0.79\%$ in the test group and $86.9 \pm 0.80\%$ in the control group, showing a difference with a p-value of 0.1, indicating it was not statistically significant.

During the second sampling, a significant difference in SY was noted: the test group had a SY of $86.5 \pm 2.19\%$, compared to $82.4 \pm 1.26\%$ in the control group, with this difference being statistically significant (p-value = 0.04).In Atlantic salmon, the first sampling showed a slaughter yield of $89.2 \pm 0.58\%$ for the test group, compared to $89.7 \pm 0.83\%$ for the control group, with a p-value of 0.3, indicating no significant difference. However, this test group's yield was significantly higher than a previous control group's SY of $87.9 \pm 0.95\%$, with a statistically significant p-value of 0.002.

Apart from diets, an assessment between species was also conducted. In the first sampling, there was a statistically significant difference in the slaughter yields between rainbow trout and Atlantic salmon, as indicated by a P-value of 0.002.

By the second sampling, the difference in slaughter yields between the species had increased, with a P-value of less than 0.001.
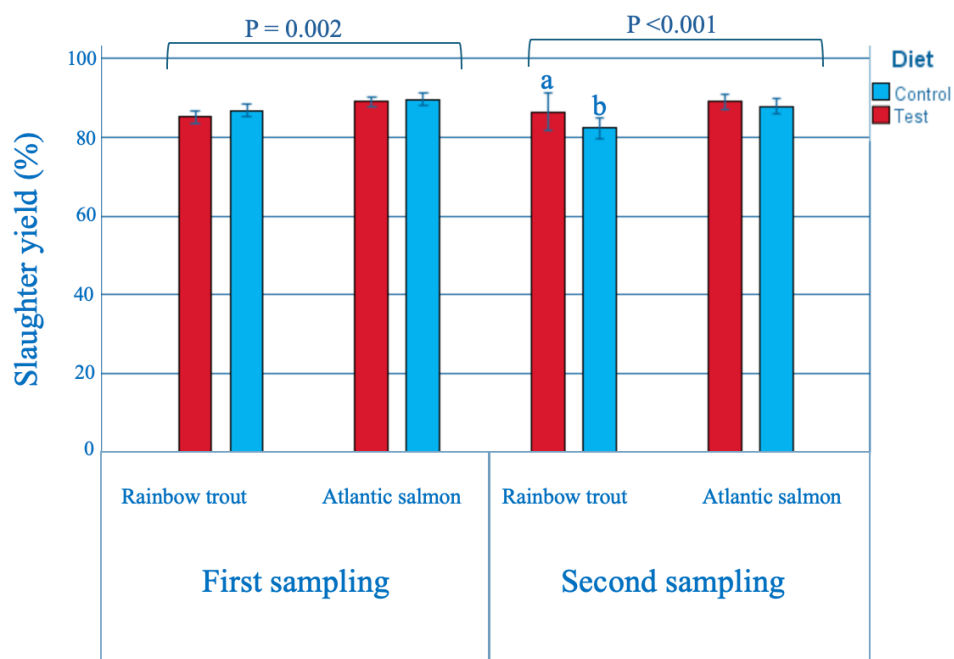

Fig17, Slaughter yield of Atlantic salmon and rainbow trout, fed control or test feed, assessed at two sampling points. The data are represented as mean values with their respective standard errors (SE). Distinct superscripts denote statistically significant differences between groups at a threshold of $p \leq 0.05$.

4.8 Fillet yield

The fillet yield of Atlantic salmon and rainbow trout, fed both control and test diets, was analyzed across two sampling times (fig,18). For rainbow trout in the first sampling, the control diet yielded a fillet of $65.58 \pm 1.22\%$, compared to the test diet's $64.83 \pm 0.79\%$. The second sampling followed a similar pattern, with the control diet yielding $64.99 \pm 1.22\%$ and the test diet slightly lower at $64.31 \pm 0.79\%$. Conversely, Atlantic salmon exhibited a different response, particularly in the second sampling. Initially, the fillet yields were almost identical, with the control group at $64.60 \pm 0.96\%$ and the test group at $64.54 \pm 0.81\%$, showing no significant difference. However, by the second sampling, a significant differences emerged; the fillet yield for the control group was $64.30 \pm 0.96\%$, while the test group decreased significantly to $62.54 \pm 0.81\%$. Apart from evaluating the effects of control and test diets, an assessment between species was also conducted. In the first sampling, the p-value is 0.13, indicating that there is no statistically significant difference between the fillet yields of the two species. In contrast, the second sampling shows a significant change with a p-value of 0.01.
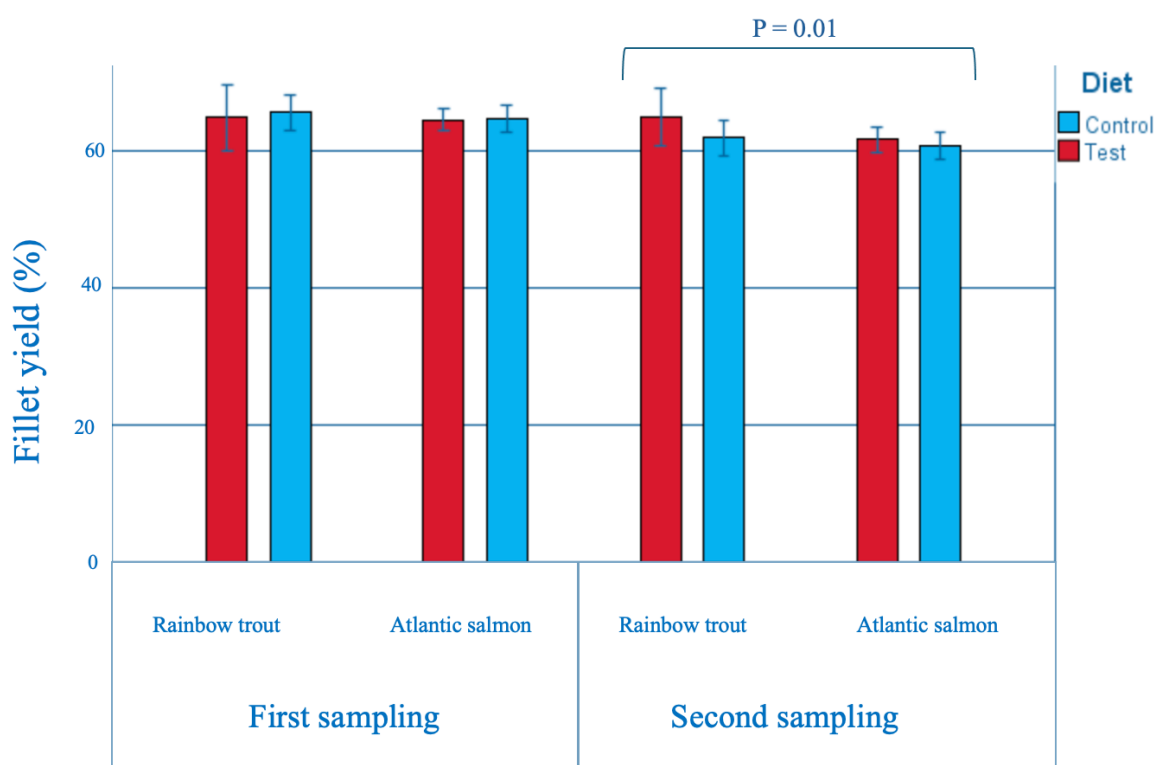

Fig18, presents the Slaughter yield of Atlantic salmon and rainbow trout, fed control and test feed, assessed at two separate sampling times. The data are represented as mean values with their respective standard errors (SE). The table also includes p-values to highlight statistically significant differences between groups at a threshold of p $\leq$ 0.05.

4.9 Fat Scores

The bar graph shows the fat scores for rainbow trout and Atlantic salmon, assessed during two sampling times.The graph highlights the differences in fat scores within each species under different dietary conditions and between the species (fig,19). In the first sampling For rainbow trout, there is no statistically significant difference in fat scores between the control and test group (P = 0.145). For Atlantic salmon, similarly, there is no significant difference in fat scores between the control and test diets (P = 0.683). In the second sampling for rainbow trout, no significant difference in fat scores is observed between the diets (P = 0.547). For Atlantic salmon, the difference between the diets also remains non-significant (P = 0.366).

Apart from evaluating the effects of control and test diets, an assessment between species was also conducted. In the first sampling there was a statistically significant difference between the species, with a P value less than 0.001. and in the second sampling, a significant difference persists between the species with a P-value less than 0.001.
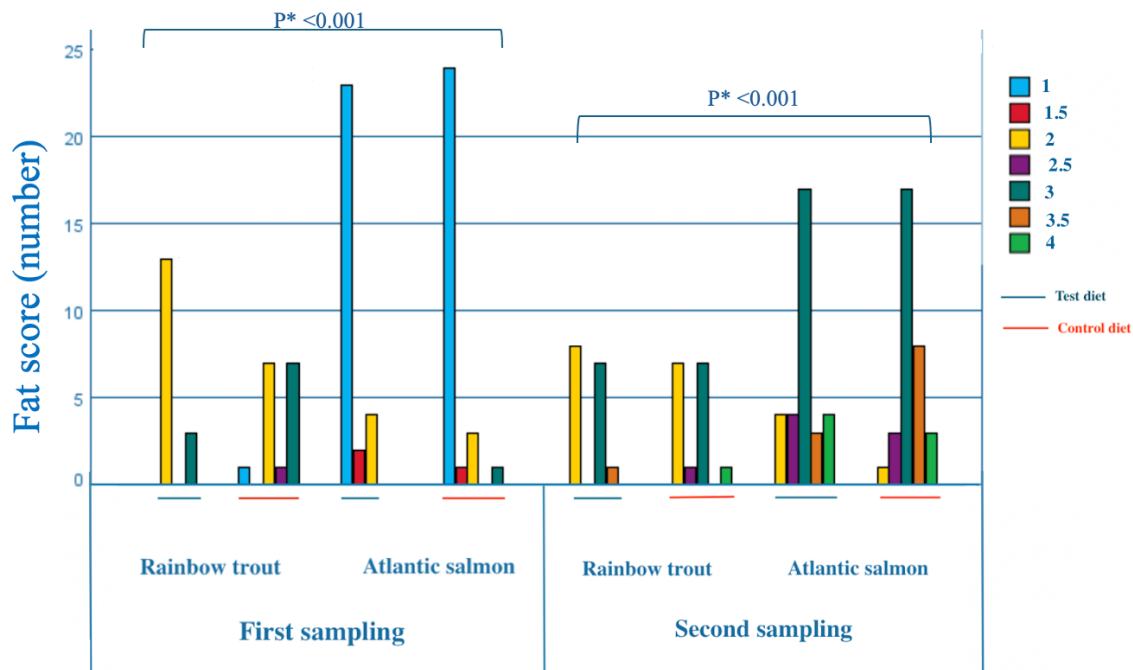

Fig19,Grouped bar chart displaying visceral fat score distribution for Rainbow troutand Atlantic salmon across two samplings with test and control diets. Chi-square analysis indicates no significant diet effect on fat deposition (p > 0.05).p* showe differences between two species.

4.10 Heart Scores

The graph illustrated the heart scores of rainbow trout and Atlantic salmon across two sampling times, comparing responses to control and test diets (fig.20). In the first sampling, rainbow trout displayed no statistically significant difference in heart scores between the control and test groups (p = 0.198). Similarly, in the second sampling, the heart scores for rainbow trout remained statistically non significant between diet groups (p = 0.6).

Atlantic salmon in the first sampling showed a statistically significant difference in heart scores between the control and test diets (p < 0.001). In the second sampling, there was no significant difference between the heart scores of the two diet groups (p = 0.274).

Apart from control and test diets, an assessment between species was also conducted. The p* value (p* < 0.001) indicated significant differences between species.

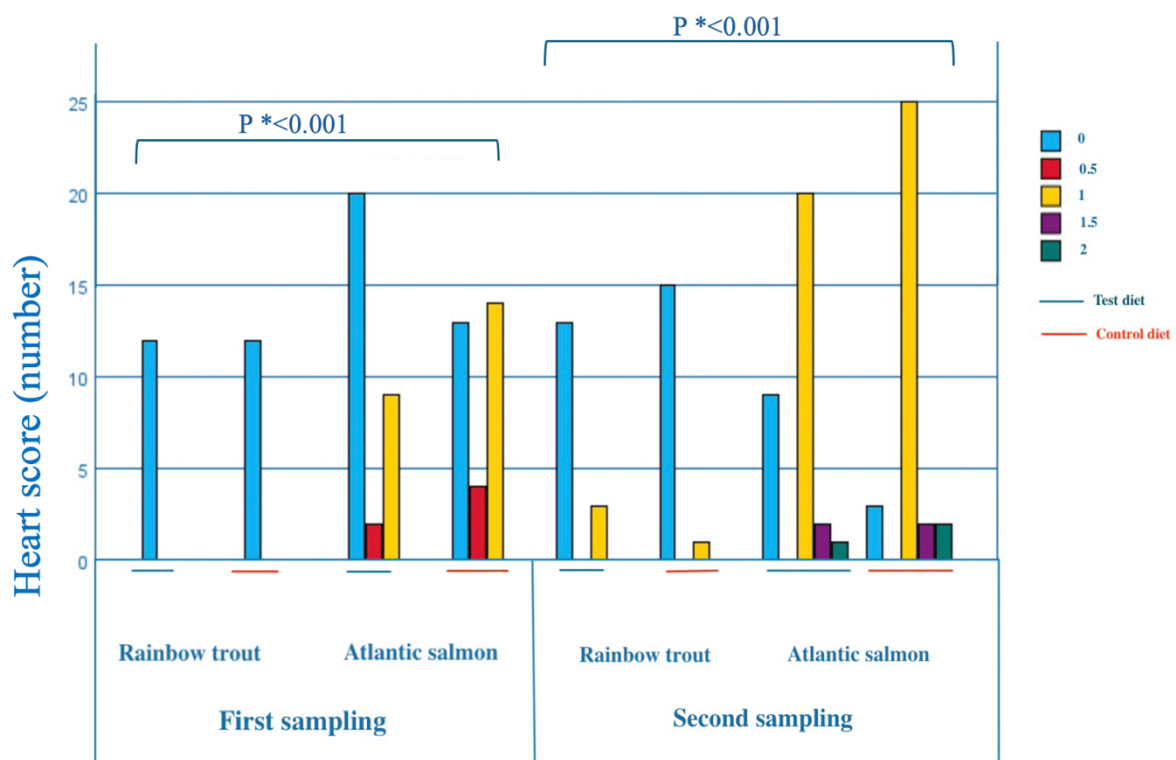*Fig20,Grouped bar chart displaying heart score distribution for Rainbow troutand Atlantic salmon on test and control diets across two sampling points, with no significant dietary effect observed (p > 0.05). p* showe differences between two species.*

4.2 Fillet quality
4.2.1 Gaping

The bar graph (fig. 21) shows gaping scores for rainbow trout and Atlantic salmon on test and control diets over two sampling periods. It evaluates how the diet affects gaping within each species and compares the differences between the species. For both sampling times, there were no significant differences in gaping scores for rainbow trout (P = 0.22 and P = 0.19) or Atlantic salmon (P = 0.29 and P = 0.19) on different diets. However, there was a significant difference between the species at the first sampling (P* = 0.04), with significant differences in gaping scores between rainbow trout and Atlantic salmon on the test diet.
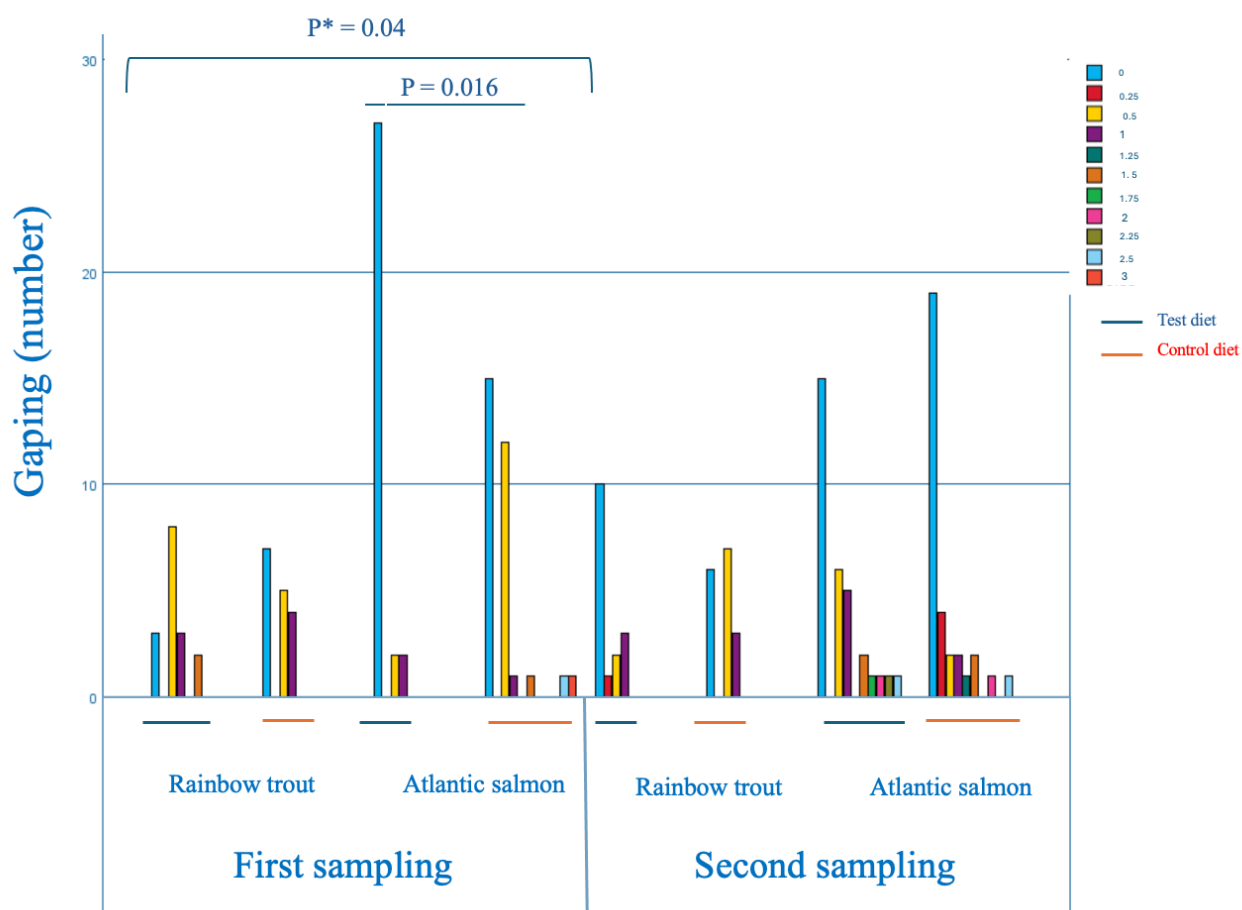

Fig21, Grouped bar chart displaying gaping score distribution for rainbow trout and Atlantic salmon on test and control diets across two sampling points significant differences between the groups (P≤0.05). p showe differences between two species.*

The bar graph (fig.22) shows the number of dark stained muscle on segments in rainbow trout and Atlantic salmon fed either the test or the control diets over two sampling times. It details how the diets affect darke segments within each species and between them.

For rainbow trout, there was no significant change in the number of dark segments between the diets at the first sampling (P = 0.31). Similarly, no significant difference emerged in the second sampling, with no dark segments observed in either the control or test groups.

For Atlantic salmon, there were significant differences in the number of dark segments during the first sampling (P = 0.01) between the control and test groups. However, in the second sampling, no significant differences were observed between the two diet groups. Despite this, the number of dark segments increased in both groups compared to the first sampling.

Additionally, when comparing the species, a significant initial difference was noted in the first sampling (P* = 0.01). By the second sampling, this difference had decreased (P* = 0.07). Over time, the number of dark segments tended to be higher in Atlantic salmon.
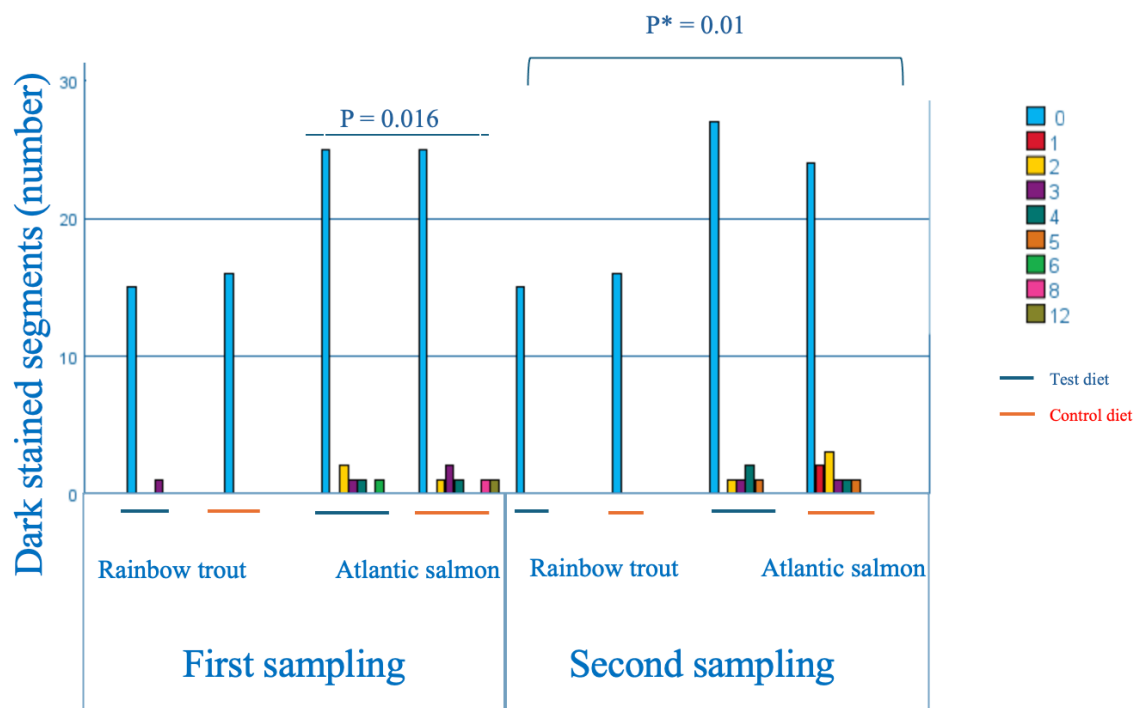

Fig22, Grouped bar chart displaying dark stained segments number distribution for rainbow trout and Atlantic salmon on test or control diets across two sampling points significant differences between the groups (P≤0.05). p* showe differences between rainbow trout and Atlantic salmon.

5. Discussion
5.1 Biometric traits

The results of this study suggest that adding 5% hydrolyzed poultry meal to the diets of Atlantic salmon and rainbow trout did not significantly affect body weight and length between the test and control groups. However, a significant difference in body weight between the two species was observed in the first sampling. In the second sampling, there were no significant differences in body weight and length between the test and control groups for both species. While body weight showed no significant difference between species, body length differed significantly. These findings are consistent with previous studies, which reported that replacing 50% of protein with poultry by-product meal in Atlantic salmon for 8 weeks (Hatlen et al., 2015)

The results showed no significant difference in the condition factor between the control and test groups of Atlantic salmon and rainbow trout during the first sampling. However, the second sampling revealed a significant difference between the test and control groups of rainbow trout (P = 0.01). Additionally, there was a significant difference between the species in both groups and both samplings. These findings align with previous studies indicating that replacing 50% of protein with poultry by-product meal in Atlantic salmon (Hatlen et al., 2015). did not affect the condition factor. The significant differences between species in both samplings might be due to temperature and species differences.

The study found that adding 5% hydrolyzed poultry meal to the diets of both Atlantic salmon and rainbow trout did not significantly affect slaughter yield between the test and control groups during the first sampling. However, in the second sampling, there was a significant difference in slaughter yield for rainbow trout (P = 0.041), but not for Atlantic salmon (P = 0.331). Additionally, there was a significant difference between the species in both groups and samplings. These results are consistent with previous studies, such as one by Hatlen et al. (2013), which reported no significant effect on slaughter yield in Atlantic salmon when comparing a diet containing European animal by-products and salmon oil with a control diet based on fish and plant ingredients (Hatlen et al., 2015). The observed differences between species could be due to variations in temperature and inherent species differences.

The study found no significant differences hepatosomatic index (HSI) between test and control diet groups in rainbow trout or Atlantic salmon in both samplings (p-values > 0.1). This suggests that the dietary variations did not significantly affect liver size in either species within the study's timeframe. However, there were consistent differences in HSI between rainbow trout and Atlantic salmon (p < 0.001), indicating distinct physiological responses between the species. These findings emphasize the importance of tailoring aquaculture practices to the specific needs of each species for optimal health and growth. Further research is needed to explore these species-specific differences and improve dietary strategies in aquaculture for enhanced sustainability and productivity.

In this study, the cardiosomatic index was examined in Atlantic salmon and rainbow trout to assess the effects of the two diets over time. For rainbow trout, both first and second samplings showed no significant differences in CSI between test and control groups, indicating stability or adaptability to dietary changes without cardiovascular stress (p-values > 0.583). In contrast, Atlantic salmon exhibited a different response: while the first sampling showed no significant difference, the second sampling revealed a significant increase in CSI in the test diet group (p = 0.029), suggesting potential metabolic or physiological adaptations. Inter-species comparisons highlighted significant differences in CSI between rainbow trout and Atlantic salmon (p < 0.001), indicating species-specific responses to dietary inputs. Understanding these differences is crucial for optimizing fish health and growth. Future research should investigate the long-term effects of dietary impacts on cardiovascular health and overall fish welfare, informing refined aquaculture practices for improved sustainability and productivity.

5.2 Fillet quality

The results indicated that gaping score in either rainbow trout or Atlantic salmon were not significantly affected by dietary changes. This suggests that gaping behavior, likely linked to muscle quality and integrity, was stable against dietary variations within the scope of the study. However, a significant species-specific difference was observed during the first sampling (P* = 0.04), where varying gaping scores between the species on the test diet were recorded. This difference was not evident in the second sampling (P* = 0.2), suggesting that an adaptation or acclimatization over time had occurred.

Regarding dark stained , no significant differences were initially shown in rainbow trout, but a noticeable change was observed in the second sampling (P = 0.01), indicating that the effects

of diet on black spots might have required time to manifest. Furthermore, significant differences were initially observed under the test diet at the first sampling ($P^* = 0.01$) when comparisons were made between species, highlighting a pronounced dietary response in one species. However, this difference diminished by the second sampling ($P^* = 0.07$), pointing to a reduction in the dietary impact over time.

These observations highlighted the complex effects of diet on physical traits in aquatic species. Gaping scores and black spot occurrences, crucial indicators of fish quality, responded variably to dietary changes across species and over time. Rainbow trout were more responsive to dietary changes in terms of black spot development, while gaping responses remained stable.

In contrast, Atlantic salmon showed minimal changes in both traits, underscoring species-specific differences in metabolism or dietary adaptability.

5.3 Chemical analyses
5.3.1 Fat Content

The assessment of fat content in rainbow trout and Atlantic salmon across two sampling periods revealed slight but statistically non-significant differences between the species.

During the first sampling, rainbow trout exhibited a fat content of 13 ± 1.03 %, numerically higher than the 10.4 ± 0.73% recorded for Atlantic salmon. However, the difference was not statistically significant (p-value = 0.06). Similarly, in the second sampling, although rainbow trout showed an increase in fat content to 14.6 ± 0.52 % compared to Atlantic salmon's 13.04 ± 0.52%, this difference also resulted in a non-significant p-value of 0.1. However, no significant differences were observed between control and test diets in either rainbow trout or Atlantic salmon. In conclusion the analysis suggests that there is no substantial variation in the fat content between rainbow trout and Atlantic salmon across the different samplings. The minor differences observed did not reach statistical significance, implying that both species may have similar fat accumulation patterns under the tested conditions. This finding could suggest that environmental factors, dietary intake, or genetic predispositions influencing fat metabolism are comparably effective across these species, at least in the context of the conditions and time frames studied. This consistency in fat content levels is crucial for aquaculture practices, indicating that similar feeding strategies could be employed for both species regarding fat intake and management.

5.3.2 Astaxanthin

The analysis of astaxanthin content in rainbow trout and Atlantic salmon across two distinct sampling periods revealed consistently higher levels in rainbow trout. In the first sampling, rainbow trout exhibited a significantly higher mean astaxanthin of 10.4 ± 0.21 compared to Atlantic salmon's 3.2 ± 0.16 mg/kg, a disparity underscored by a highly significant p-value of less than 0.001. This trend persisted into the second sampling, where rainbow trout maintained higher astaxanthin (12.2 ± 0.41) relative to Atlantic salmon, which exhibited an increase to 6 ± 0.4 mg/kg. However, no significant differences were observed between control and test diets in either rainbow trout or Atlantic salmon.

In conclusion the findings strongly suggest that rainbow trout naturally accumulates higher levels of astaxanthin compared to Atlantic salmon under the conditions studied. The significant differences noted, particularly in the first sampling, reinforce the notion of inherent metabolic or genetic differences between these species that influence how they assimilate and store astaxanthin.

5.3.3 Fillet Fatty Acid Composition

The study of fatty acid profiles in Atlantic salmon and rainbow trout revealed how each species uniquely responds to changes in diet. These insights highlight the differences in their metabolic processes and the implications for aquaculture diet formulations.

Atlantic salmon showed little change in fatty acid levels when diets were altered, suggesting a stable metabolism that is less sensitive to dietary changes. However, there were significant decreases in the monounsaturated fatty acid C22:1n11 and increases in the polyunsaturated fatty acid C20:2 with the test diet, indicating that some specific fatty acids can still be affected by targeted dietary changes.

Rainbow trout, on the other hand, responded more dynamically to dietary changes, showing significant increases in the saturated fatty acid C14:0 and decreases in the monounsaturated fatty acid C16:1n7. This suggests that rainbow trout have a more adaptive metabolism that could be exploited to enhance certain fatty acids beneficial for aquaculture.

The comparison between the species showed notable differences; rainbow trout had higher levels of several important fatty acids, while Atlantic salmon had higher levels of C20:1 and C20:2. This emphasizes the species-specific nature of fatty acid metabolism, which could be due to genetic factors or evolutionary adaptations.

These results are valuable for refining aquaculture diets to optimize fish health and growth. They also suggest that rainbow trout's sensitivity to dietary changes could make them a useful model for researching dietary impacts on fatty acid metabolism in other species.

5.3.4 Heart Fatty Acid Composition

Significant variations were noted in $C20:4n6$ levels across both control and test groups, as well as in $C18:0$ in the test groups between species. These findings are consistent with previous research on fatty acid composition in both wild and farmed Atlantic salmon and rainbow trout. Although the previous study by Blanchet et al. (2005) indicated no significant differences in fatty acid profiles between wild and farmed fish, it observed that farmed Atlantic salmon typically had higher levels of $C20:4n6$, corroborating the results of the current study (Blanchet et al., 2005). Another investigation by Belghit et al. (2019) examined the effects of a diet based on black soldier fly larvae in Atlantic salmon and found that while the diet was rich in unsaturated fatty acids, it did not significantly alter the fatty acid composition of the fish's whole body or tissues (Belghit et al., 2019) . Polyunsaturated fatty acids generally exhibit higher concentrations in the heart compared to other tissues (Skuladottir et al., 1990). Additionally, Liu et al. (2019) reported that lower environmental temperatures lead to increased levels of unsaturated fatty acids in phospholipids across all tissues, although the predominant fatty acids in both control and test feeds were monounsaturated fatty acids (Liu et al., 2019).

5.4 Myocommata width

Initial results indicated that the myocommata width differences between the test and control diets in rainbow trout were not significant (p = 0.77). Similarly, Atlantic salmon showed consistent myocommata widths across both diets (p = 0.93). In the second set of measurements, the widths for the control diet in both Atlantic salmon and rainbow trout showed no significant changes (p = 0.2 and p = 0.3, respectively).

Despite the non-significant dietary effects within each species, the comparative analysis between species revealed a different story. Statistically significant differences were found in the myocommata widths between rainbow trout and Atlantic salmon, with values significantly varying (p = 0.03 in the first sampling and p = 0.001 in the second sampling). This indicates a marked disparity in muscle structure development influenced possibly by genetic or physiological differences rather than dietary intake.

Further, the study analyzed the correlation between fork length and myocommata width, highlighted through scatter plots (fig10). For rainbow trout, a moderate positive correlation was demonstrated (R² = 0.4331), suggesting that around 43.31% of the variation in myocommata width could be explained by changes in fork length. In contrast, Atlantic salmon exhibited a stronger correlation (R² = 0.7249), indicating that about 72.49% of the variability in myocommata width was attributable to variations in fork length. This higher correlation in Atlantic salmon suggests a more pronounced influence of body size on muscle structure compared to rainbow trout.

These findings suggest that while dietary formulations can be optimized, understanding species-specific physiological and genetic influences on growth and body structure is crucial for effective aquaculture management. Future studies should continue to explore these biological variations and their implications for fish farming practices to optimize growth and health outcomes based on species-specific needs.

5.8 Fat Score

For rainbow trout, there was no significant difference in fat scores between the control and test diets (P = 0.1), indicating that the diet did not significantly affect fat accumulation. Similarly, Atlantic salmon showed no significant difference in fat scores between the control and test diets (P = 0.6), suggesting a similar resilience to dietary changes.

In the second sampling period, both species continued to show no significant differences in fat scores between the control and test diets. Rainbow trout had a P-value of 0.5, and Atlantic salmon had a P-value of 0.3, indicating that dietary changes did not significantly affect fat accumulation in either species.

This result contrasts with previous studies, such as Bell et al. (2010), which reported that diet significantly influenced visceral fat content in Atlantic salmon fed vegetable oil (Bell et al., 2010).When comparing fat scores between rainbow trout and Atlantic salmon, a significant difference was observed in both sampling periods, with P-values of 0.001. This indicates that species-specific physiological differences contribute to variations in fat scores, regardless of diet.In conclusion, this analysis shows that both rainbow trout and Atlantic salmon are resilient to dietary changes in terms of fat accumulation.

5.9 Heart Score

Results indicated that there were no significant differences between the test and control groups within species during the first sampling. However, by the second sampling, significant differences emerged between the test and control groups in Atlantic salmon. Differences between species were also significant in both samplings. The larger fish in the second sampling, which had spent more time in seawater, were expected to show more fat deposition on heart than wild fish (Poppe et al., 2003).

Further research into the effects of poultry meal and porcine by-product meal on Atlantic salmon showed no notable lesions when compared to controls (Liland et al., 2015). Additionally, another study found that a diet including soybean fermented with 4% macro algae sugar kelp (Saccharina latissima) did not significantly impact fat accumulation around the heart, although it was noted to lower the heart score in Atlantic salmon in both samplings (Formanowicz, 2022).

For rainbow trout, there was no change in heart score in the first sampling, aligning with findings from Renna (2017) where adding up to 40% partially defatted black soldier fly larvae to the diet did not affect the species. However, a higher heart score was observed in the second sampling due to dietary differences. The observed significant differences between species in both samplings likely reflect inherent biological differences (Renna et al., 2017).

6. Conclusion

The results indicate that the inclusion of chicken hydrolysate in the diet of rainbow trout leads to significant differences, particularly showing reductions in visceral fat and gutted weight, while increasing the slaughter yield. However, the fat score was not significantly different in Atlantic salmon and rainbow trout based on the diets, although significant differences were observed between the two species; notably, Atlantic salmon exhibited significantly higher visceral fat score, which increased over time.

Myocommata width did not show significant changes based on the diet; however, it did increase over time within each species and was consistently higher in Atlantic salmon. Interestingly, no significant correlation was found between myocommata width and gaping.

In terms of the fatty acid profiles in the fillets, all fatty acids were significantly higher in rainbow trout, except for C20:1 and C20:2. Additionally, a higher deposition of EPA and DHA was noted in the fillets of rainbow trout, whereas in Atlantic salmon, these were more prominently deposited in the heart tissue. This differential deposition highlights species-specific responses to the diet.

As rainbow trout tended to have higher fat content in muscle compared with salmon, but at the same time less visible fat content, the result showed that rainbow trout and Atlantic salmon store the fat differently in the muscle.

Slaughter yield in Atlantic salmon was significantly higher than in rainbow trout at both sampling points. However, the lower slaughter yield in rainbow trout indicates a higher mass of visceral fat compared to Atlantic salmon.

Visceral fat accumulation increased over time in Atlantic salmon. Similarly, the score for fat accumulation on the heart surface was significantly higher in Atlantic salmon over time. In the second sampling, the control group of Atlantic salmon had significantly higher fat accumulation on the heart surface compared to the test group.

Hypothesis	Yes/No
Does the inclusion of poultry hydrolysate in the diet affect the fat deposition patterns in both Atlantic salmon and rainbow trout?	No
There are species-specific responses to poultry hydrolysate supplementation, affecting biometric traits, fat deposition patterns, and fillet quality in Atlantic salmon and rainbow trout.	Yes

Refrences

Albrektsen, S., Kortet, R., Skov, P. V., Ytteborg, E., Gitlesen, S., Kleinegris, D., Mydland, L. T., Hansen, J. Ø., Lock, E. J., & Mørkøre, T. (2022). Future feed resources in sustainable salmonid production: A review. *Reviews in aquaculture*, *14*(4), 1790-1812.

Andersen, U. B., Strømsnes, A. N., & Steinsholt, K. (1994). Norwegian Journal of Agricultural Sciences: fillet gaping in farmed Atlantic salmon (Salmo Salar).

Belghit, I., Waagbø, R., Lock, E. J., & Liland, N. S. (2019). Insect-based diets high in lauric acid reduce liver lipids in freshwater Atlantic salmon. *Aquaculture Nutrition*, *25*(2), 343-357.

Bell, J. G., Pratoomyot, J., Strachan, F., Henderson, R. J., Fontanillas, R., Hebard, A., Guy, D. R., Hunter, D., & Tocher, D. R. (2010). Growth, flesh adiposity and fatty acid composition of Atlantic salmon (Salmo salar) families with contrasting flesh adiposity: Effects of replacement of dietary fish oil with vegetable oils. *Aquaculture*, *306*(1-4), 225-232.

Bell, J. G., & Waagbø, R. (2008). Safe and nutritious aquaculture produce: benefits and risks of alternative sustainable aquafeeds. *Aquaculture in the Ecosystem*, 185-225.

Blanchet, C., Lucas, M., Julien, P., Morin, R., Gingras, S., & Dewailly, É. (2005). Fatty acid composition of wild and farmed Atlantic salmon (Salmo salar) and rainbow trout (Oncorhynchus mykiss). *Lipids*, *40*(5), 529-531.

Boyd, C. E., McNevin, A. A., & Davis, R. P. (2022). The contribution of fisheries and aquaculture to the global protein supply. *Food security*, *14*(3), 805-827.

Cadillo-Benalcazar, J. J., Giampietro, M., Bukkens, S. G., & Strand, R. (2020). Multi-scale integrated evaluation of the sustainability of large-scale use of alternative feeds in salmon aquaculture. *Journal of Cleaner Production*, *248*, 119210.

Einen, O., Waagan, B., & Thomassen, M. S. (1998). Starvation prior to slaughter in Atlantic salmon (Salmo salar): I. Effects on weight loss, body shape, slaughter-and fillet-yield, proximate and fatty acid composition. *Aquaculture*, *166*(1-2), 85-104.

Fallah-Delavar, M., & Farmani, J. (2018). Recovery and characterization of enzymatic protein hydrolyzates and fat from chicken skin. *Journal of the American Oil Chemists' Society*, *95*(9), 1151-1161.

FAO. (2020). World Fisheries and Aquaculture. *Food and Agriculture Organization*, *2020*, 1-244.

FAO. (2022). *FAO Fisheries and Aquaculture Division*. FAO. https://doi.org/https://doi.org/10.4060/cb8609en

Fiskeri- og kystdepartementet, F. (2009). *Strategy for an Environmentally Sustainable Norwegian Aquaculture Industry*. UNEP Law and Environment Assistance Platform Retrieved from http://www.regjeringen.no/fkd

Fiskeridir. (2021). *fiskeridir*. https://www.fiskeridir.no/English/Aquaculture/Statistics/Booklets/_/attachment/download/8afd87c7-68ed-4d8d-9d47-1b13c5c28186:1b843760204b861b789fac3773fda48385a255dc/nokkeltall-havbruk-2021-eng.pdf

Formanowicz, J. (2022). *Product quality, fish welfare and salmon louse infestation of Atlantic salmon and rainbow trout fed a diet supplemented with fermented soybean and macro algae (Saccharina latissima)* Norwegian University of Life Sciences, Ås].

Galkanda-Arachchige, H. S., Wilson, A. E., & Davis, D. A. (2020). Success of fishmeal replacement through poultry by-product meal in aquaculture feed formulations: a meta-analysis. *Reviews in aquaculture*, *12*(3), 1624-1636.

Hatlen, B., Jakobsen, J. V., Crampton, V., Alm, M., Langmyhr, E., Espe, M., Hevrøy, E., Torstensen, B., Liland, N., & Waagbø, R. (2015). Growth, feed utilization and endocrine responses in Atlantic salmon (Salmo salar) fed diets added poultry by-product meal and blood meal in combination with poultry oil. *Aquaculture Nutrition*, *21*(5), 714-725.

Hekmatpour, F., Kochanian, P., Marammazi, J. G., Zakeri, M., & Mousavi, S.-M. (2018). Inclusion of poultry by-product meal in the diet of Sparidentex hasta: Effects on production performance, digestibility and nutrient retention. *Animal Feed Science and Technology*, *241*, 173-183.

Hodar, A., Vasava, R., Mahavadiya, D., & Joshi, N. (2020). Fish meal and fish oil replacement for aqua feed formulation by using alternative sources: a review. *Journal of Experimental Zoology India*, *23*(1).

Hua, K., Cobcroft, J. M., Cole, A., Condon, K., Jerry, D. R., Mangott, A., Praeger, C., Vucko, M. J., Zeng, C., & Zenger, K. (2019). The future of aquatic protein: implications for protein sources in aquaculture diets. *One Earth*, *1*(3), 316-329.

Hurdalsplattformen. (2021). https://www.regjeringen.no/contentassets/cb0adb6c6fee428caa81bd5b339501b0/no/pdfs/hurdalsplattformen.pdf

Jacobsen, A., Joensen, H., & Eysturskarð, J. (2017). Gaping and loss of fillet firmness in farmed salmon (Salmo salar L.) closely correlated with post-slaughter cleaning of the abdominal cavity. *Aquaculture Research*, *48*(1), 321-331.

Jacobsen, A. s., Shi, X., Shao, C., Eysturskarð, J., Mikalsen, S.-O., & Zaia, J. (2019). Characterization of glycosaminoglycans in gaping and intact connective tissues of farmed Atlantic Salmon (Salmo salar) fillets by mass spectrometry. *ACS omega*, *4*(13), 15337-15347.

Jobling, M. (2001). Nutrient partitioning and the influence of feed composition on body composition. *Food intake in fish*, *25*(4), 354-375.

Liland, N., Hatlen, B., Takle, H., Venegas, C., Espe, M., Torstensen, B., & Waagbø, R. (2015). Including processed poultry and porcine by-products in diets high in plant ingredients reduced liver TAG in A tlantic salmon, S almo salar L. *Aquaculture Nutrition*, *21*(5), 655-669.

Lim, C., Lee, C.-S., & Webster, C. D. (2023). *Alternative protein sources in aquaculture diets*. CRC Press.

Lindland, K. M., Gjerstad, B., Krøvel, A. V., & Ravagnan, E. (2019). Governing for sustainability in the Norwegian aquaculture industry. *Ocean & Coastal Management*, *179*, 104827.

Liu, C., Dong, S., Zhou, Y., Shi, K., Pan, Z., Sun, D., & Gao, Q. (2019). Temperature-dependent fatty acid composition change of phospholipid in steelhead trout (Oncorhynchus mykiss) tissues. *Journal of Ocean University of China*, *18*, 519-527.

Mørkøre, T., Moreno, H. M., Borderías, J., Larsson, T., Hellberg, H., Hatlen, B., Romarheim, O. H., Ruyter, B., Lazado, C. C., & Jiménez-Guerrero, R. (2020). Dietary inclusion of Antarctic krill meal during the finishing feed period improves health and fillet quality of Atlantic salmon (Salmo salar L.). *British Journal of Nutrition*, *124*(4), 418-431.

Mørkøre, T., & Rørvik, K.-A. (2001). Seasonal variations in growth, feed utilisation and product quality of farmed Atlantic salmon (Salmo salar) transferred to seawater as 0+ smolts or 1+ smolts. *Aquaculture*, *199*(1-2), 145-157.

Myhre, M. R., Roger. (2022). *Analyse marint restråstoff 2021* (SINTEF Ocean rapporter;2022:00501 A, Issue. S. O. AS. https://hdl.handle.net/11250/3013196

Naylor, R. L., Hardy, R. W., Bureau, D. P., Chiu, A., Elliott, M., Farrell, A. P., Forster, I., Gatlin, D. M., Goldburg, R. J., & Hua, K. (2009). Feeding aquaculture in an era of finite resources. *Proceedings of the National Academy of Sciences*, *106*(36), 15103-15110.

Naylor., Hardy, R. W., Bureau, D. P., Chiu, A., Elliott, M., Farrell, A. P., Forster, I., Gatlin, D. M., Goldburg, R. J., Hua, K., & Nichols, P. D. (2009). Feeding aquaculture in an era of finite resources. *Proceedings of the National Academy of Sciences*, *106*(36), 15103-15110. https://doi.org/doi:10.1073/pnas.0905235106

NCE. (2022). Future ingredients

for Norwegian

salmon feed.

Nengas, I., Alexis, M., & Davies, S. (1999). High inclusion levels of poultry meals and related byproducts in diets for gilthead seabream Sparus aurata L.[Pagrus aurata].

O'Fallon, J. V., Busboom, J., Nelson, M., & Gaskins, C. (2007). A direct method for fatty acid methyl ester synthesis: Application to wet meat tissues, oils, and feedstuffs. *Journal of animal science*, *85*(6), 1511-1521.

Pelletier, N., Klinger, D. H., Sims, N. A., Yoshioka, J.-R., & Kittinger, J. N. (2018). Nutritional attributes, substitutability, scalability, and environmental intensity of an illustrative subset of current and future protein sources for aquaculture feeds: joint consideration of potential synergies and trade-offs. *Environmental science & technology*, *52*(10), 5532-5544.

Pittman, K., Grigory, V., & Brandebourg, T. (2013). Bridging the gap to sustainable salmon farming: overcoming the gaping problem. *Journal of Fisheries & Livestock Production*, *1*(2).

Poppe, T. T., Johansen, R., Gunnes, G., & Tørud, B. (2003). Heart morphology in wild and farmed Atlantic salmon Salmo salar and rainbow trout Oncorhynchus mykiss. *Diseases of Aquatic Organisms*, *57*(1-2), 103-108.

Rahnama, B. (2010). *Supplementing a commercial diet for atlantic salmon (Salmo salar L.) with arginine, glutamate or tetradecylthioacetic acid (TTA). impact on production efficiency, slaughter parameters and flesh quality*

Regjeringen. (2022). *Regjeringen*. Retrieved from https://www.regjeringen.no/no/tema/forskning/innsiktsartikler/langtidsplanen-for-forskning-og-hoyere-utdanning-2023-2032/id2929453/?expand=factbox2930882

Regulation, E. (2009). laying down health rules as regards animal by-products and derived products not intended for human consumption and repealing Regulation (EC) No 1774/2002 (Animal by-products Regulation). . *Official Journal of the European Union, Brussels, Belgium.* .

Renna, M., Schiavone, A., Gai, F., Dabbou, S., Lussiana, C., Malfatto, V., Prearo, M., Capucchio, M. T., Biasato, I., & Biasibetti, E. (2017). Evaluation of the suitability of a partially defatted black soldier fly (Hermetia illucens L.) larvae meal as ingredient for rainbow trout (Oncorhynchus mykiss Walbaum) diets. *Journal of animal science and biotechnology*, *8*, 1-13.

Rouger, A., Tresse, O., & Zagorec, M. (2017). Bacterial contaminants of poultry meat: sources, species, and dynamics. *Microorganisms*, *5*(3), 50.

Shepherd, C. J., Monroig, O., & Tocher, D. R. (2017). Future availability of raw materials for salmon feeds and supply chain implications: The case of Scottish farmed salmon. *Aquaculture*, *467*, 49-62.

Skuladottir, G., Schiöth, H., Gudmundsdottir, E., Richards, B., Gardarsson, F., & Jonsson, L. (1990). Fatty acid composition of muscle, heart and liver lipids in Atlantic salmon, Salmo salar, at extremely low environmental temperature. *Aquaculture*, *84*(1), 71-80.

Steffens, W. (1994). Replacing fish meal with poultry by-product meal in diets for rainbow trout, Oncorhynchus mykiss. *Aquaculture*, *124*(1-4), 27-34.

Tacon, A. G., & Metian, M. (2008). Global overview on the use of fish meal and fish oil in industrially compounded aquafeeds: Trends and future prospects. *Aquaculture*, *285*(1-4), 146-158.

Villamil, O., Váquiro, H., & Solanilla, J. F. (2017). Fish viscera protein hydrolysates: Production, potential applications and functional and bioactive properties. *Food Chemistry*, *224*, 160-171.

Wang, W. (2016). *The effect of dietary antioxidants on hyperpigmented fillet spots of Atlantic salmon (Salmo salar L.)* Norwegian University of Life Sciences, Ås].