



Greenhouse gas emissions of rainbow trout fed conventional and novel feeds from Baltic region, evaluated using Life Cycle Assessment

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#### Mistra Food Futures Report #13 Greenhouse gas emissions of rainbow trout fed conventional and novel feeds from the Baltic region evaluated using life cycle assessment. Utsläpp av växthusgaser från regnbåge som utfodras med konventionellt och nytt foder från Östersjöregionen, utvärderat med hjälp av en analys av den totala livscykeln.

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The overarching vision of the programme Mistra Food Futures is to create a science-based platform to enable transformation of the Swedish food system into one that is sustainable (in all three dimensions: environmental, economic and social), resilient and delivers healthy diets. By taking a holistic perspective and addressing issues related to agriculture and food production, as well as processing, consumption and retail, Mistra Food Futures aims to play a key role in initiating an evidence based sustainability (including environmental, economic and social dimensions) and resilience transformation of the Swedish food system. This report is a part of Mistra Food Future's work to identify agricultural systems with potential to make agriculture net-zero, one of the central issues within Mistra Food Futures.

Mistra Food Futures is a transdisciplinary consortium where key scientific perspectives are combined and integrated, and where the scientific process is developed in close collaboration with non-academic partners from all parts of the food system. Core consortium partners are Swedish University of Agricultural Sciences (SLU), Stockholm Resilience Centre at Stockholm University and RISE Research Institutes of Sweden.

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### Abstract

Aquaculture production set a new record in 2020, with over 120 million tonnes of production, which corresponds to about half of the global seafood consumption. However, Swedish aquaculture production is currently low, but slowly increasing. The global aquaculture sector is predicted to continue to grow but needs to reduce its environmental footprint. In intensive aquaculture in which feed is used, feed inputs often account for the largest share of environmental impacts, thus feed development is a priority to increase the sustainability of fed aquaculture.

The purpose of this study is to evaluate the environmental sustainability implications of shifting to more regional and circular feed inputs for rainbow trout, by, as a first step, estimating the greenhouse gas emissions – or carbon footprint- of the novel feed and fish raised on it compared to conventional production. Fish were produced in net pens in Sweden and fed either a conventional feed (reference), or an experimental feed in which 60% of the protein content derives from novel ingredients (insects, blue mussels, sea squirts and fava bean protein isolate) sourced from the Nordic countries to replace land animal by-products (i.e. blood meal and poultry by product meal) and soy protein concentrate.

Results show that the novel feed reduces greenhouse gas emissions of one kg of rainbow trout by around 63 %. Fish fed the experimental feed maintained the same growth and economic feed conversion ratio (eFCR) as fish fed the control feed. The reduction is mainly due to the almost 70% lower emissions of the experimental feed; 1.6 kg CO<sub>2</sub>eq./kg feed compared to 5.4 kg CO<sub>2</sub>eq./kg feed of the conventional feed. Feeding fish insects reared on plant-based waste streams from the food industry, increases the circularity and reduces emissions. However, the modelling choice that some feed inputs based on side streams with no economic value are free of environmental burden, has a strong influence on the results. Despite shorter transport distances no lower impact of transports could be found for the experimental feed due to the utilisation of more climate intensive transport means/modes. Further, the novel feed ingredients used in the study come from pilot or test scale production plants, with potential to further decrease emissions with optimised processing. At present, the available volumes of these feed inputs are limited which prevents a rapid large-scale shift of the aquaculture industry. Other sources of uncertainty include the fact that the FCR is based on a four-month growth trial which might not reflect a complete production cycle. This study indicates that there is a potential to reduce the carbon footprint of intensive aquaculture by using alternative protein sources, an important step that shows that it is worthwhile to continue expanding the analysis to cover also other environmental aspects to avoid shifting burdens between different types of environmental impact.

Keywords: Rainbow trout, aquaculture, LCA, feed, greenhouse gas emissions, novel proteins.

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# 1. Introduction

The aquaculture production reached a record in 2020, with 122.6 million tonnes (live weight equivalent), and provides about half of the global seafood supply and is predicted to continue grow. Only the global salmonid aquaculture has in the last 30 years grown from 0.5 to 3.8 million tons of annual production, with industries in Norway and Chile as the leading producers (FAO, 2022). Seafood consumption is on average 12 kg edible seafood per person and year in Sweden, corresponding to approximately 1-2 meals per week (Hornborg et al., 2021), which is lower than the recommendation from the Swedish Food Agency. The most common type of seafood consumed in Sweden is imported from Norway: farmed Atlantic salmon (Salmo salar), and to a smaller extent farmed rainbow trout (Oncorhynchus mykiss). Swedish consumers state that they want to buy more seafood produced in Sweden, and they think that seafood produced in Sweden is more sustainable compared to imported products (Johansson and Skog, 2015). Further, seafood production in Sweden is currently low, as is the theoretical degree of Swedish seafood self-sufficiency (Hornborg et al., 2021), although the potential in especially aquaculture in Sweden is high. The environmental footprint of seafood differs greatly between products, where seaweed and bivalves generate the lowest environmental pressure, and farming of crustaceans and flatfish the highest (Gephart et al., 2021). However, except for green house gas emission from aquaculture production, which is fairly well known (Macleod et al., 2020), less is known of other sustainability impact categories like land or freshwater use and results between studies are often hard to compare due to incompatible methodologies (Gephart et al., 2021). To reduce the environmental footprint of aquaculture, feed development is a priority since feed inputs most often account for the largest share of environmental impacts (Bohnes et al., 2019; Pelletier et al., 2009).

As carnivorous species, salmonids have a high dietary requirement for proteins and highquality amino acids. Further, to fully utilize the growth potential, most salmonids require an energy dense diet, with a high content of poly unsaturated fatty acids (i.e. omega-3) (National Research Council, 2011). The feed for salmonids is to a large extent composed of high-quality protein ingredients and lipids, formerly of marine origin. As many fish stocks are already exploited at, or above, their maximum capacity, efforts have been made to decouple aquaculture from these limited resources. Thus, modern aquafeeds for intensive production of carnivorous fish has developed from being heavily dependent on marine input, to consist of 75% plant proteins and oils, to a high degree imported to Europe where the feed production takes place (Aas et al., 2019; Johansen et al., 2022).

The growing aquaculture require more feed raw materials, and especially in the salmon industry much attention are given novel feed ingredients, as these are thought to enable a more sustainable growth, while meeting the requirements of both fish and consumer (Albrektsen et al., 2022; Colombo et al., 2022). Insect larvae, with special focus on black soldier fly (Hermetia illucens) and mealworms (Tenebrio molitor), are potential sources of protein which has gained increasing attention for their potential use in fish feed (Nogales-Mérida et al., 2019). Insect rearing requires comparatively little space and energy, and the nutrient requirements of the insects are, as far as we know, quite low (allowing the use of food industry side streams like e.g. potato peel) and the feed is efficiently converted to biomass due to being ectothermic animals (Thévenot et al., 2018). Blue mussels (Mytilus edulis) and sea squirts (Ciona intestinalis) represents other interesting feed alternatives for salmonids. Blue mussels and sea squirts can be farmed without external inputs of feed or fertilizers, and their cultivation has a relatively small environmental impact on a mass basis, and the nutritional composition could offer an interesting alternative source of protein (Albrektsen et al., 2022). Their extractive nature can even help mitigate local eutrophication. However, there are knowledge gaps when it comes to their function as fish feed, especially in combination with each other. Further, in order to know if these novel developments are steps towards more sustainable aquaculture feeds, their environmental performance needs to be evaluated in a transparent and science-based way.

The integration of aquaculture in the circularity bioeconomy framework (reviewed by Colombo et al., 2022), where waste from one system is reused as input in another system, has been practiced for decades in the form of by-products of animal and plant origin and (D'Abramo and Ziegler, 2022) even for longer in integrated extensive pond culture. The transformation of modern aquafeeds, to a be part of circular bioeconomy requires a conceptual change in thinking, shifting focus from increasing productivity to increased resource use efficiency. An adoption to this approach is a strategic and a resilient way forward for the aquaculture industry, which is vital to reduce its environmental footprint. Moreover, the complexity and fragility of raw materials and product supply chain of aquaculture feeds for the European market, was highlighted during the COVID-19 pandemic and recent geo-political instability in Eastern Europe, as well as the obstruction of the Suez Canal in 2021 (Colombo et al., 2022).

The concept behind this study is the development of a sustainable aquaculture feed with novel protein sources, based on circular nutrient flows in the Baltic region. In the study, the environmental performance of shifting to a more circular feed is assessed by quantifying greenhouse gas emissions – or carbon footprint- of farmed rainbow trout fed an experimental feed based on ingredients from the Nordic countries, compared to a conventional feed, in a feeding trial where the fish was raised to slaughter size.

# 2. Methods

#### 2.1.1. Goal and scope

The goal of this study is to evaluate the environmental sustainability implications of shifting to a more regional and circular feed inputs for rainbow trout. This is done by estimating of the greenhouse gas (GHG) emissions of rainbow trout produced in net pens in Sweden, fed either a conventional (reference) feed, or the experimental feed, in which 60% of the protein content derives from novel ingredients (i.e. insects, blue mussel, sea squirt meal, fava bean protein isolate) sourced from the Nordic countries. Further, GHG emissions from the individual novel feed raw materials are estimated and hotspots in the production are identified. Production of feed raw materials, feed composition and production, and fish trial was performed in the Vinnova financed project "fem ton grön fisk i disk" (grant: 2019-00854). The standardised LCA methodology, according to the International Organisation of Standardization; 14040 and 14044 (ISO, 2006a and 20006b), is used. The intended audience is wide and ranges from fish farmers and feed producers to downstream customers of the products, including wholesalers and retailers. The findings can also be of interest for seafood certifying organisations and policy makers.

### 2.1.2. Functional unit

In this study two functional units are used which represent different stages in the production of rainbow trout in aquaculture systems. The first functional unit considered is 1 of kg rainbow trout feed at the feed production plant gate for both the reference and experimental feed formulation. Additionally, the impact of head on gutted (HOG) rainbow trout was analysed using the functional unit 1 kg of head on gutted rainbow trout, at the processing plant gate.

#### 2.1.3. System boundaries

The system boundaries of the rainbow trout production include the production (fishing, agriculture or other and their primary processing to meals, oils and protein concentrates) and transport of feed ingredients, transport of feed to the farm, material and energy use during rearing in a net pen, transport to processing plant and subsequent processing (Fig. 1). Feed use of fry and juveniles, until start of the feeding trial, was included. However, as the energy and material use connected to hatchery, rearing of fry and juvenile production

were the same for the two cases to be compared, and due to the general low importance (Johansen et al., 2022) of these inputs in combination with data unavailability, it was decided to exclude them. This exclusion lead to an expected minor underestimation of total results, and focus should therefore be on relative results rather than absolute.

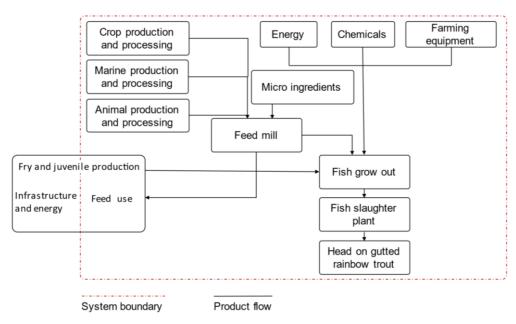


Figure 1. System boundaries of the studied rainbow trout production supply chain.

## 2.1.4. Allocation

This study uses mass allocation in cases of co-production following the hierarchy of allocation methods presented in ISO 14040 and ISO 14044 (2006a and 20006b). Co-products, i.e., products that have an economic value for the producer and are further utilized in another supply chain, share the same footprint per unit of product. Co-products that do not have an economic value were not assigned any impact. This is further detailed under section 2.2 for the specific feed ingredients and products.

## 2.1.5. Impact assessment

Characterisation factors from IPCC (2021), 100-year time horizon, were used to calculate the global warming potential (GWP). Background data was sourced from Ecoinvent 3 (Version 3.8) and the Agri-footprint database (Version 5.0). The LCA model was built in SimaPro Developer Multi User (version 9.4.).

# 2.2. Life cycle inventory

### 2.2.1. Feed formulations

During the growth trial, fish were fed either a reference feed or the experimental feed (Table 2). Ingredient composition of the reference feed, as well as origin, was provided by the producer. The test feed consisted of protein sources from insect larvae mealworm and black soldier fly reared on food waste, marine inputs from Atlantic herring (*Clupea harengus*) and European sprat (*Sprattus sprattus*) fished in the Baltic Sea, and meals from the sea squirt and blue mussel cultured in the Skagerrak, and fava bean protein isolate (originated from Norway). The experimental feed did not include any soy products. Both feeds used in the trial fulfilled the nutrient requirements of rainbow trout, and had similar crude protein, lipid and energy content (Table 1).

INGREDIENT (%)	REFERENCE FEED	EXPERIMENTAL FEED	
FISHMEAL	15.0	12.0	
MUSSEL MEAL	-	3.0	
SEA SQUIRT MEAL	-	4.0	
FISH OIL	7.0	13.0	
PROCESSED ANIMAL PROTEINS <sup>1</sup>	16.0	-	
BLACK SOLDIER FLY	-	12.5	
MEALWORMS	-	16.5	
CROP BASED PROTEINS <sup>2</sup>	12.0	12.0	
CARBOHYDRATES/STARCH <sup>3</sup>	20.0	11.0	
RAPESEED OIL	19.0	13.5	
MICRO INGREDIENTS <sup>4</sup>	4.0	2.6	
REUSE FEED	7.0	-	
NUTRIENT COMPOSITION (%)			
CRUDE PROTEIN	41.0	40.6	
CRUDE LIPID	31.0	31.0	

Table 1. Ingredient and nutrient compositions of reference and experimental diets

<sup>1</sup>Blood and poultry by-product meal.

<sup>2</sup>Fava bean protein isolate, soy protein concentrate, corn gluten and wheat gluten.

<sup>3</sup>Wheat meal and fava bean.

<sup>4</sup>Includes vitamin and mineral premixes, amino acids, astaxanthin and other feed additives.

## 2.2.2. Novel feed ingredients

The impact data sources for the feed ingredients used in the feeds in this study are a combination of primary and secondary data (Table 2). Black soldier fly (BSF), mealworms, mussel meal, fish meal and oil are based on primary data.

INGREDIENT (%)	PRIMARY DATA	DATA SOURCE	
FISHMEAL	Yes	Major European producer	
MUSSEL MEAL	Yes	Small scale Swedish producer	
SEA SQUIRT MEAL	No	(Bergentz, 2017)	
FISH OIL	Yes	Major European producer	
POULTRY BY-PRODUCT MEAL	No	Database data (modified)	
BLOOD MEAL (PORCINE)	No	Database data (modified)	
BLACK SOLDIER FLY LARVAE	Yes	SLU	
MEALWORMS	Yes	Commercial Swedish producer	
CROP BASED PROTEINS	No	Database data	
CARBOHYDRATES/STARCH	No	Database data	
RAPESEED OIL	No	Database data	
MICRO INGREDIENTS	No	Winther et al. (2020)	

Table 2. Data sources of the feed ingredients assessed in this study

#### Black soldier fly

The BSF larvae used in the experimental feed were produced by the Swedish University of Agricultural Sciences (SLU), Uppsala, Sweden. Here a container unit, specifically designed for insect larvae rearing was utilized to produce the larvae used in this study. Breeding stock and additional, rearing related activities (e.g. washing of trays, feed preparation, drying and packaging) were performed in a separate building. In the data used, material and energy use during operations for both larvae and adult stage BSF rearing and post-rearing processing were included, covering the production from cradle-to-gate. Infrastructure use of both the container unit and building were excluded. The BSF larvae were fed a mix of unsold supermarket bread and unsold vegetables from a wholesaler in Stockholm and these ingredients were shredded before being fed to the larvae. The adult flies were fed waste chicken or pig feed from SLU's research farm in Uppsala. An important methodological decision taken was the assumption that all feed ingredients used to feed BSF, both larvae and flies, represented current waste streams and where therefore considered free of environmental burden. This builds on the assumption that these ingredients would have been thrown away otherwise and serve no alternative use in the current, local food and feed system. Obviously, waste streams should be minimized, but those that do occur now and, in the future, should be used as efficiently as possible.

#### Mealworm

Mealworm larvae used in the production of the experimental feed assessed in this study were produced by a small, but commercial scale, Swedish company. The larvae were fed a mixture of potato peels, wheat bran and yeast. Potato peels are a waste stream from the food industry, i.e. it has no economic value, and were, following the same argumentation as in the case of BSF larvae, considered free of environmental burden. Both wheat bran and yeast are commercially used feed ingredients and therefore carried full environmental burden. All production was located at the same locality and energy use for heating (wood chip based) and electricity use were included. Additionally, material use of rearing boxes, cleaning materials and packaging was included while infrastructure use of the building was excluded. The company assessed sells the larvae faeces, also called "frass", as a fertilizer and the environmental impact of production was therefore divided between the larvae and the frass based on output mass. The system boundaries applied to the assessment of this ingredient were cradle-to-grate.

#### Fishmeal and fish oil

Fishmeal and fish oil used in the experimental and reference feeds were produced by a Finnish company, using herring and sprat from Finnish pelagic fisheries in the Baltic Sea. For the fishery, fuel use data of Baltic reduction fisheries (Hornborg, in prep.) was used to accurately reflect the species mix and associated fuel cost of these locally sourced marine ingredients. For processing, i.e., fish reduction to meal and oil, data from a major European fishmeal producer was used as a proxy. Material use during production (e.g. cleaning agents and additives) was included but packaging was excluded as fishmeal and oil are typically shipped in bulk. The production was modelled from cradle-to-factory gate.

#### Mussel meal

Mussel meal is based on blue mussels which are cultivated on long-lines on the Swedish west coast and processed to meal by a small-scale company. The production line utilised electricity when running the machinery used for deshelling and grinding the mussels and heating oil was utilised when drying the wet mass to obtain the final product. Data collection of in and outputs for this processing line is based on estimations by the producer and is connected to uncertainty. This producer operated a pilot scale when providing mussel meal for the experimental feed and has since upgraded to a more efficient processing line.

# 2.2.3. Transports

Transports happened through all parts of feed production and later rainbow trout grow out and processing. The transport distances of the single feed ingredients for both feeds were calculated using Google Maps<sup>1</sup> and seadistances.org<sup>2</sup> (Table 3). Inner Swedish transport of the complete feeds from the feed mill to the grow out site was assumed to be 400 km. Transport of table sized fish to the slaughter and processing plant was assumed to be 100 km.

EXPERIMENTAL FEED INGREDIENT	ORIGIN	DISTANCE TRUCK (KM)	DISTANCE FERRY (KM)	DISTANCE BOAT (KM)
FISHMEAL	Finland	650	150	-
FISH OIL	Finland	650	50	-
WHEAT GLUTEN	Sweden	400	-	-
WHEAT MEAL	Sweden	400	-	-
FAVA BEAN ISOLATE	Lithuania	400	-	-
RAPESEED OIL	Sweden	400	-	-
MEALWORM MEAL	Sweden	730	-	-
BSF MEAL	Sweden	470	-	-
SEA SQUIRT MEAL	Sweden	220	-	-
MUSSEL MEAL	Sweden	220	-	-
VITAMIN MINERAL PREMIX	Asia	50	-	23 150
DL-METHIONINE	Asia	50	-	23 150
MONOCALCIUM PHOSPHATE	Asia	50	-	23 150
VIT-C35	Asia	50	-	23 150
ASTAXANTHIN (NATURAL, BACTERIAL)	UK	50	-	1667
REFERENDE FEED INGREDIENT				
FISHMEAL	Finland	300	-	-
FISH OIL	Finland	300	-	-
WHEAT GLUTEN	Finland	300	-	-
CORN GLUTEN	Germany	1240	200	-
FAVA BEAN	Finland	300	-	-
SOY PROTEIN CONCENTRATE	Brasil	50	-	12 038
WHEAT MEAL	Finland	300	-	-
POULTRY BY-PRODUCT MEAL	Finland	300	-	-
BLOODMEAL (PORCINE)	Germany	1240	200	-
RAPESEED OIL	Finland	300		-
VITAMINS, MINERALS AND OTHER ADDITIVES	Global market mix	50	-	23 150
AMINO ACIDS	Global market mix	50	-	1945
REUSE FEED	In house	N/A	N/A	N/A

**Table 3.** Origin, transport mode and distances of the individual feed ingredients used in bothassessed feeds

<sup>&</sup>lt;sup>1</sup> www.google.com/maps

<sup>&</sup>lt;sup>2</sup> https://sea-distances.org/

## 2.2.4. Feed production

The experimental feed was produced at the Center for Feed Technology at the Norwegian University of Life Sciences, Ås, whereas Raisio AB in Finland produced the reference feed. The energy and material inputs required for feed production were however modelled independent of location and based on Winther et al. (2020) which focussed on the production of Atlantic salmon feed, which are similar in both composition and production method to rainbow trout feed.

# 2.2.5. Fry and juvenile production

Feed use of the fish before the start of the experiment (i.e. until 1.1 kg) was included in the assessment under the simplified assumption that the reference and experimental feeds were fed during the whole lifecycle of the produced fish. Further the eFCRs gained during the feed trial were assumed to be applicable for the whole lifecycle as well. All other inputs into the hatchery, fry and juvenile production, until start of the feeding trial, were not considered in this study, including material and energy use, as described under system boundaries. Information about the production method for the trout used in the study was unavailable and other studies have shown an only marginal influence of these inputs to the total carbon footprint of salmonids raised in aquaculture systems (Johansen et al., 2022; Winther et al., 2020).

## 2.2.6. Grow out operations and fish processing

The feed experiment was carried out at a commercial aquaculture site located in Dalarna using open net pens in freshwater (see section 2.3). For modelling, data from Finnish rainbow trout production was used as production technology and scale are comparable to Swedish production (Silvenius et al., 2012). Fish were slaughtered and gutted before being sold to restaurants and consumers. Energy and material use of processing operations were modelled following Winther et al. (2020) and a rainbow trout specific conversion factor of 1.215 from head on gutted (HOG) to liveweight was used (Norwegian Directorate of Fisheries, 2018). However, all emissions from the rainbow trout farming are allocated to the product, since it was assumed that guts from slaughtering were not further utilized.

# 2.3. Grow out trial

The fish trial was carried out at Älvdalslax (production site in Österdalsälven, Dalarna, Sweden) for about four months in 2021, with start in June. In total 2040 rainbow trout (average weight of 1.11 kg) were transferred to two smaller net pens, where the feeding trial started. The fish where slaughtered in October, with an average final weight of 2.39 kg for fish fed reference feed and 2.37 kg for fish fed the test feed. No mortalities were recorded in any of the two net pens during the trial. The economic feed conversation ratio (eFCR) was calculated according to the following equation:

 $eFCR = \frac{given \ feed \ (kg \ feed)}{biomass \ end \ (kg \ live \ weight) - biomass \ start(kg \ live \ weight)}$ 

The feed conversation ratio for fish consuming reference feed and test feed were 1.01 and 1.05, respectively.

# 3. Results

# 3.1. Novel feed ingredients

### 3.1.1. Black soldier fly

Black soldier fly larvae, like many insect larvae, tolerate a range of conditions but thrive best at stable temperatures and humidity levels. Electricity use of the air conditioning unit in the grow out container unit stands for a majority of emissions of this ingredient followed by the electricity use during drying. When ready to ship out as a feed ingredient (dried and packaged product) the BSF larvae have GHG emissions of 0.8 kg CO<sub>2</sub> eq./kg.

### 3.1.2. Mealworm

Mealworms require similar conditions to BSF larvae but the use of biofuels to heat the rearing facilities reduces the contribution of heating to the total carbon footprint to <10%. Electricity use of the machinery, lighting and other rearing related activities accounts for about 40% of the GHG emissions of mealworms. The highest share is being taken by the two commercial feed ingredients, wheat bran and yeast, which together account for about 50% of the total emissions. The dried and packaged end product has GHG emissions of 1.5 kg  $CO_2$  eq./kg.

### 3.1.3. Fishmeal and oil

The raw material for the fishmeal and oil used in this study are Baltic sprat and herring, typically caught using pelagic trawls. The emissions associated with this fishery, predominately stemming from the burned fuel, stand for three quarters of the total emissions of the fishmeal and oil. The remaining quarter is divided between the energy inputs in fishmeal processing and feed additives. At factory gate the fishmeal and oil have GHG emissions of 2.0 kg  $CO_2$  eq./kg.

## 3.1.4. Mussel meal

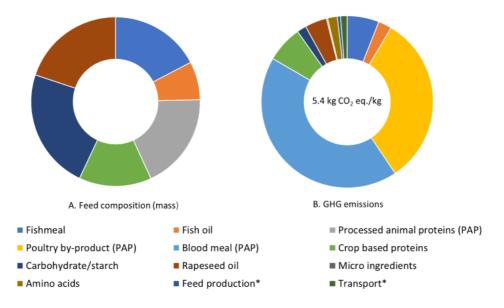
When providing blue mussel meal for the experimental feed, the producer operated using a pilot scale production line which had not been energy optimised and the data must be considered to have high uncertainty. The farming of the blue mussels used as raw material account for about 20% of the total GHG of the finished mussel meal. An emission hot spot in the production line was the drying step, which utilised heat from burned heating oil to dry the ground mussel flesh into a meal. The heating oil consumed and burned here accounted for about 75% of the total emissions. Electricity use only played a marginal role. The final, dried product has a GHG of  $6.2 \text{ kg CO}_2 \text{ eq./kg}$ .

# 3.2. Climate impact of feeds

#### 3.2.1. Reference feed

When summarizing emissions of the ingredients, transportation and feed production, the reference feed emits 5.4 kg CO<sub>2</sub> eq./kg feed. Processed animal proteins (PAP) (i.e. poultry by products and blood meal) dominate emissions in particular in relation to their inclusion rates (Fig. 2). Feed inputs that have a higher inclusion, but lower relative contribution to feed emissions are rapeseed oil, carbohydrates, amino acids, fishmeal and the crop-based proteins. The content of crop-based proteins is 12%, corresponding to 7% of emissions, while inclusion of 18% PAPs corresponds to 76% of the GHG emissions of the product. Energy use in feed production and transportation in the reference feed give small contributions to emissions.

A key factor leading to the comparatively high GWP of the reference feed are emissions from land use change (LUC), which are 30% of total emissions. These predominately originate from the PAPs as these are based on pig or poultry rearing, which in turn requires feed which typically includes ingredients connected to LUC (e.g. soy beans cultured in countries with expanding agricultural land). The use of soy protein concentrate (SPC) is another ingredient connected to LUC.



*Figure 2. Relative contribution to mass (A) and greenhouse gas emissions (B) of different ingredients, feed production and transports in the reference feed (\*only in B).* 

### 3.2.2. Experimental feed

Emissions of the experimental feed are  $1.6 \text{ kg CO}_2$  eq./kg feed, delivered to the fish farm, i.e. almost 70% lower than the reference feed. The ingredients contribute more proportionally to emissions which means they are more even in emission rates (Fig. 3). The protein ingredients that give a higher contribution relative to inclusion rate are mussel meal and fish meal and those that give a lower contribution than their inclusion rates are sea squirt meal, insect meals, fava bean isolate and wheat gluten. The major contributors to GHG emissions in the experimental feed are fish oil, mealworms and fish meal (17%, 16% and 15%, respectively), but these ingredients do also stand for high inclusion rates (42% in mass). Noteworthy is the mussel meal and astaxanthin that together contributes to 15% of the GHG emissions, but only 3% in mass. The experimental feed is not greatly influenced by LUC derived emissions (<5%). This is based on the direct avoidance of ingredients connected to LUC e.g. by replacing SPC with fava beans as well as the phasing out of PAPs through the use of insects, fed on local crops. The relative contribution of feed production and transports to the GHG emissions of the experimental feed, are higher than for the reference diet, due to the lower emissions of the feed ingredients. The actual numbers for feed production are the same for the two feeds. The emissions of transports for the different feeds reveals a slightly higher impact for the experimental feed (0.02 kg)CO<sub>2</sub> eq./kg feed) than the corresponding value for the reference feed, despite shorter transport distances, implying more climate intensive transport means are used which more or less cancels out the effect of shorter distance

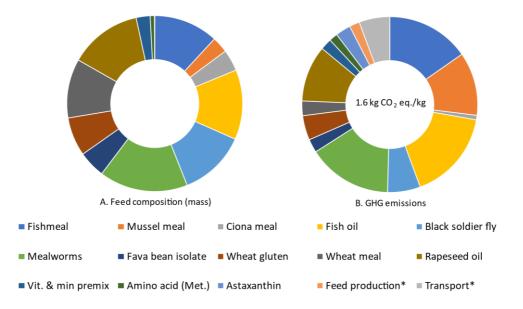
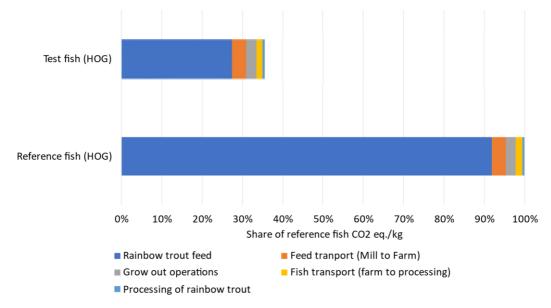


Figure 3. Relative contribution to mass (A) and greenhouse gas emissions (B) of different ingredients, feed production and transports in the test feed (\*only in B).

# 3.3. Climate impact of the fish

Feed production constituted 78% of total emissions of the experimental trout and 92% of the reference trout. The experimentally fed rainbow trout had 64% lower emissions compared to the conventionally fed trout (Fig. 4). With very similar eFCRs for fish fed both the reference and experimental feeds, it can be concluded that the experimental feed was well-balanced and made the fish grow equally as the reference feed and that the difference in emissions caused by feed can be largely attributed to the ingredients used. As grow out related operations, as well as transports and processing of fish are independent of feed ingredient sourcing, there is no difference in emissions stemming from these sources between the final products based on the reference and experimental feeds.



*Figure 4.* Relative greenhouse gas emissions of rainbow trout (head on gutted (HOG)) at the processing plant gate.

# 4. Discussion

In the present study, conventional aquaculture feed ingredients, with a relatively high carbon footprint, like soy protein concentrates and animal proteins (i.e. blood meal and poultry by product meal), were replaced with novel protein sources with a lower carbon footprint. The result shows that this can successfully be performed without a reduction in productivity (i.e. fish growth) and with a decreased carbon footprint of both feed and final product, rainbow trout. Compared with the average ingredient composition in feed for Atlantic salmon in Norway for 2021 (Johansen et al., 2022), both the reference and test feeds in this study has a lower inclusion of crop-based ingredients (experimental: 37%; reference: 51%; Norwegian salmon: 71%) and differs in the inclusion rate of marine feed inputs (32%; 19% and 29%, respectively). However, excluding novel marine protein sources in the test feed (i.e. blue mussel and sea squirt), the content of fishmeal and -oil was 25% which is in line with the content in conventional salmon feeds the since the last 10 years (Aas et al., 2019; Johansen et al., 2022).

Almost half of the crude protein content in the experimental feed originates from insects reared on plant-based waste streams from the food industry. Black soldier fly larvae and mealworms have successfully been evaluated as fish feed components in several studies and reviews (Caimi et al., 2021; Cardinaletti et al., 2019; English et al., 2021; Nogales-Mérida et al., 2019; Renna et al., 2017), and have been pointed out as a future local/regional protein source for the Norwegian salmon industry (Albrektsen et al., 2022; Almås et al., 2020; Skogli et al., 2022). However, the processing (e.g., defatting, drying etc.) and chemical composition of insect meals differs between studies, as well as formulation of experimental feeds (e.g. higher fish meal content than in commercial feeds) which brings in uncertainties in the interpretation of the results. Tran et al. (2022) performed a metaanalysis of production performance of fish fed insect meals and concluded that dietary intake of mealworm larvae in fish supports growth, in contrast to several other insect species including BSF larvae, in comparison to fish meal-based control diets. Further, the authors concluded that insect meal holds a great potential to but needs to be integrated with other novel and conventional protein sources. In another meta-analysis (Liland et al., 2021) it was concluded that there is a maximum threshold of 25-30% inclusion rate of insects in feeds for farmed fish, without a negative impact on fish performance. In the present study, mealworms and black soldier fly larvae were combined with other novel protein sources in a low fishmeal and fish oil diet, and completely replaced soy and animal by-products,

without any observed depression in growth or increased feed conversion ratio. The latter is of high importance to maintain a low environmental footprint of the product.

In the trial, the FCR was similar and the feed emissions 1.6 and 5.4 CO<sub>2</sub> eq./kg experimental and reference feed, respectively. If the FCR for fish the experimental feed had been higher, this difference would be reduced. To reduce the GHG impact of aquaculture feeds, the total exclusion of fish meal and oil and instead replace it with land PAPs and soy protein concentrate will rather have the opposite effect. Parker (2017) presented the correlation between carbon footprint of salmon feed and input of animal by-products (ABP), and even though there is some inconsistency between studies on the emissions from ABP (e.g. Dekamin et al., 2015; Pelletier et al., 2009; Ytrestøyl et al., 2011), the conclusion was that GHG emissions increase with inclusion of ABP in the feed. In general, emissions from poultry by-products have lower GHG emission than land production of mammals, in particular ruminants, and the allocation method (e.g. allocation based on economic value or impact not allocated to by-products) are both factors affecting the estimated impact from ABP. In addition to the lower environmental footprint of reduction fisheries (Cashion et al., 2017), fish meal and -oil are excellent sources of highly digestible essential nutrients, as well as important for the palatability of fish feed.

The calculated GHG emissions of the rainbow trout presented in this study, is based on a four-month growth period and the assumption that FCR values were the same during the whole life span until slaughter. The FCR is a function of growth, which differs during the life span of the fish (Austreng et al., 1987). Data on the growth and feed use during the whole production cycle would give more accurate data. In addition, for a better precision more fish and several pens should be included. Further, all novel feed ingredients utilised in the experimental feed assessed in this study come from pilot or test scale production plants, with temporary energy sources and equipment. This comes with challenges to both representativity and data quality as these operations often operate using non-optimised methods, producing small volumes and sometimes do not have the instruments needed for accurate accounting of all in- and outputs. Especially insect production is a new and developing area and there is no industry wide utilised "best technology". Neither is the processing of mussels to a meal, which includes deshelling, drying and grinding, which is a complex process to industrialize. The production methods used in the rearing of both insect species used in the test feed are therefore not necessarily representative of the entire industry. However, since the data collection in this study was completed, multiple producers have started expanding and optimising their production and are in the process of scaling up their production, opening the opportunity for larger available volumes of novel feed ingredients in the future.

One core decision which influenced the results of this study greatly is the modelling choice that some, direct or indirect, feed inputs based on side streams are considered free of charge (e.g. potato peel, waste vegetables and bread) whereas other side streams that have an economic value (e.g. pig blood used for bloodmeal production) carried their share of environmental burden. The underlying reasoning is that products which in the current food system already are a product of economic value are considered a product from the production system just as the main product, and which therefore also should have an environmental cost connected to it. If a side stream is utilised which (in the current system) is of no value, this product is considered free of environmental burden as it would rather require further investment of money and energy to dispose of if not used in feed production. If disregarding whether a side stream currently has an economic value or not, i.e. either considering all side stream free from environmental burden or if they would carry their full environmental burden, the assessed feeds and fish would be closer in carbon footprint. The economic value of side stream can obviously also change over time, which would influence the modelling.

One main objective within the "Fem ton grön fisk i disk" project is the creation of a feed which not only focusses on locally or regionally sourced ingredients but also increasing circularity in the Swedish food production system. Therefore, a focus on ingredients like insects, which utilise waste streams were chosen. Baltic fishmeal and oil, blue mussels and sea squirts assimilate nutrients, and close nutrient loops within the watershed, and, while not being side streams, are therefore regarded as circular in a larger nutrient flow perspective. The focus on carbon footprint in this study does not capture the potential benefits of nutrient recycling and defining methods to assess circularity as part of or complementing LCA studies would be beneficial. An improvement potential to this study would be the inclusion of further impact categories focusing on nutrient emissions (e.g. eutrophication). Like circularity, biodiversity loss is a key factor not captured by this assessment. LCA methods to assess biodiversity loss are currently under development for both marine and terrestrial biodiversity but no established method has been implemented to broader LCA assessments as of now. Biodiversity loss is a key challenge in our current global challenges and in some cases highly connected to climate change. Expanding the scope of this study to include a measure of biodiversity loss, when such a measure and data to assess it become available is therefore a key improvement potential. Further, despite shorter transport distances no lower impact of transports could be found for the experimental feed due to the utilisation of more climate intensive transport means/modes. This is in line with data from Modahl and Brekke (2022), who concluded that transport of insect meals from Central and Western Europe to the west coast of Norway for feed production makes only a minor contribution to the environmental burdens.

Another challenge connected to the pilot-scale production of novel feed ingredients are the available quantities for feed production. Current production volumes of insects, sea squirtand mussel meals, as well as fava bean protein isolate, are low and would not be sufficient to replace more traditional feed ingredients in fish feeds used in Swedish aquaculture in any larger scale.

# 5. Conclusions

The result from this study shows that it is possible to formulate a functioning rainbow trout feed based on Nordic ingredients while focussing on circular and sustainable choices. The trout fed with the experimental feed showed satisfactory growth. Replacing feed inputs connected to land use change like animal-based by-products and soy with low-input and regionally produced feed inputs led to major emission reductions. However, more local is not by definition more sustainable, how each feed input is produced and transported determines its sustainability. Similarly, less marine inputs do not always lead to lower emissions, it depends on how these are produced and what they are replaced by. Here, marine inputs from the region were shown to be a relatively low emission feed input providing important nutrients. Despite uncertainties both in data and methodological assumptions, the novel feed was shown to have a major potential to reduce emissions of farmed trout. This study could in the future be extended to include other, alternative Nordic ingredients and a more extensive fish trial covering the entire lifecycle of the fish as well as cover additional types of environmental impact.

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