Sustainable farming of Arctic charr
(*Salvelinus alpinus*)

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Abstract
Arctic charr (Salvelinus alpinus L.) is a salmonid that is well adapted to cold waters and is commonly farmed in open net cages. A breeding program has enabled a relatively large production in Sweden, providing a continuous supply of fresh fish to the market. Despite expansions and improvements of the Arctic charr farming industry during the last decade, there are still sustainability challenges to overcome.

This doctoral thesis explores various methods for improving the sustainability of Arctic charr farming, such as alternative protein sources in the feed, a shortened production cycle through selective breeding and an adaptive feeding management model.

A new feed composition, containing a protein mixture with ingredients that are not attractive for human consumption, was evaluated for Arctic charr. The feed consisted of Baltic Sea decontaminated fishmeal (Sprattus sprattus and Clupea harengus), Baltic Sea blue mussel meal (Mytilus edulis) and baker’s yeast (Saccharomyces cerevisiae). The new feed resulted in lower growth compared with fish fed a commercial-type control feed, likely due to a reduced digestibility caused by the baker’s yeast. However, a self-selection study revealed a preference for the new feed over the control feed. Additionally, consumers could not distinguish between fish that had been fed the new feed or the control feed. Possible family effects that the new feed may have on growth were also investigated, showing that selection for high growth on a commercial-type feed would also benefit a higher growth capacity on the new feed.

Selective breeding of Arctic charr had clear positive effects on growth. After adjusting for changes of environmental factors within the hatchery, the growth improvement from the first generation to the current seventh generation was 11 % per generation and production time has been shortened by ten months. Selective breeding also affected seasonal growth patterns and Arctic charr from the seventh generation grew more during winter than previously, although the largest weight improvement between generations occurred during summer. A new growth capacity, and pattern, demands an adaptive feeding management model so that feed waste and environmental impacts can be minimized. A new model on the digestible energy need of Arctic charr and a seasonal growth capacity factor were developed and can together be used to calculate the daily feed allowance during different periods of the growth season.

Keywords: Arctic charr, Salvelinus alpinus, aquaculture, Saccharomyces cerevisiae, Mytilus edulis, breeding, feeding management, sustainability

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This thesis is based on the work contained in the following papers, referred to by Roman numerals in the text:


II Carlberg, H., Lundh, T., Cheng, K., Pickova, J., Langton, M., Varzquez Gutierrez, J., Kiessling, A., Brännäs, E. In search for protein sources: evaluating an alternative to the traditional fish feed for Arctic charr (Salvelinus alpinus L.). (Submitted manuscript).

III Carlberg, H., Alanärä, A., Brännäs, E. Evaluation of family effects on growth performance of Arctic charr (Salvelinus alpinus L.) fed an alternative feed. (Manuscript).

IV Carlberg, H., Nilsson, J., Brännäs E., Alanärä, A. An evaluation of 30 years of selective breeding in Arctic charr (Salvelinus alpinus L.) and its implications for feeding management. (Manuscript).

Paper I is reproduced with the permission of the publishers.
The contribution of H. Carlberg to the papers included in this thesis was as follows:

I  Performed the experiment and wrote parts of the manuscript.

II  Performed the experiments, samplings, parts of the chemical analyses, the main part of the statistical analyses, and wrote the main part of the manuscript.

III  Performed the experiment, statistical analyses, calculations, and wrote the manuscript.

IV  Collected and analysed large parts of the data and wrote the main part of the manuscript.
# Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
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<tbody>
<tr>
<td>ADC</td>
<td>Apparent digestibility coefficient</td>
</tr>
<tr>
<td>CL</td>
<td>Crude lipid</td>
</tr>
<tr>
<td>CP</td>
<td>Crude protein</td>
</tr>
<tr>
<td>DE</td>
<td>Digestible energy content of the feed</td>
</tr>
<tr>
<td>DEN</td>
<td>Digestible energy need</td>
</tr>
<tr>
<td>DM</td>
<td>Dry matter</td>
</tr>
<tr>
<td>FA</td>
<td>Feed allowance</td>
</tr>
<tr>
<td>SGR</td>
<td>Specific growth rate</td>
</tr>
<tr>
<td>TER</td>
<td>Theoretical energy requirement</td>
</tr>
<tr>
<td>TGC</td>
<td>Thermal growth coefficient</td>
</tr>
<tr>
<td>TW&lt;sub&gt;i&lt;/sub&gt;</td>
<td>Theoretical weight increment</td>
</tr>
</tbody>
</table>
1 Introduction

Aquaculture is a fast growing industry that has expanded greatly during the last 40 years (FAO, 2016; Ytrestøyl et al., 2015). On average, every second fish eaten globally is farmed (FAO, 2016) and fish farming creates a valuable protein source with little effort compared to many other protein producing activities (Kaushik & Médale, 1994). Along with a growing human population and new eating habits the world’s need for animal protein is increasing. In the industrialized countries, we have increased the consumption of food originating from aquatic environments markedly (FAO, 2016). In the EU, we consume 23.1 kg fish per person annually and 5.5 kg of these originate from aquaculture production (EU, 2016). Since humans push nature harder and harder, many wild fish stocks are depleted or extinct (FAO, 2016; Pauly et al., 2001).

The aquaculture industry and its development face many challenges. The industry is very diverse; many species are farmed and different methods are used. Some challenges that are often mentioned are the origin of the feed ingredients (Deutsch et al., 2007; Naylor et al., 2000), escapees of farmed fish causing negative genetic and ecological impacts (Fraser et al., 2010; Soto et al., 2001), spreading of diseases and parasites (Krkošek et al., 2007), use of antibiotics and chemicals, nutrient output from the farms (Gowen & Bradbury, 1987), animal welfare (Olesen et al., 2011; Huntingford et al., 2006), and impacts on surrounding ecosystems (Subasinghe, 2009; Naylor et al., 2000).

1.1 Aquaculture in Sweden

Swedish aquaculture is a small industry, producing 0.05 % of the total global production (EU, 2016; FAO, 2016). Farming is often conducted in open systems using net-pens or land based flow through systems in fresh or brackish water. The nutrient output from the farms has occasionally been problematic. The fish species farmed are mainly salmonids, rainbow trout (*Oncorhynchus mykiss*) and
Arctic charr (*Salvelinus alpinus* L.) are the most common species (SCB, 2015). They are often sold as rather expensive food, fresh over counter or as processed delicacy products. The Swedish aquaculture industry is growing and there is a trend towards fewer but larger farms (Sather *et al.*, 2013). Between 2007 and 2011 the tonnage of fish produced for the markets almost doubled but ever since, the production has been kept rather stable at 11 000 tones (SCB, 2015).

### 1.2 Arctic charr farming

The salmonid Arctic charr is an extreme cold water adapted fish species (Johnson, 1980) that can grow in very low temperatures (Brännäs & Wiklund, 1992). The species can be either anadromous or land-locked and has a circumpolar distribution. It is our most northerly distributed salmonid and is often found in clean and pristine high altitude mountain lakes (Johnson, 1980).

The Arctic charr farmed in Sweden is a strain from lake Hornavan that was selected in the early 1980’s to make up for the basis of the Swedish breeding program for Arctic charr; given the brand name Arctic superior (Nilsson *et al.*, 2010). The breeding program has resulted in a stable commercial production of Arctic charr and a growing interest from the market (Eriksson *et al.*, 2010).

Approximately 1700 tonnes of Arctic charr are farmed annually in Sweden, making up for 15% of the total tonnage of farmed fish in Sweden and 20% of the total value (SCB, 2015).

Farming is often conducted in net-pens in cold water bodies in the rural northern part of Sweden. These waters are usually naturally oligotrophic and often also affected by hydropower regulations (Eriksson *et al.*, 2010). Iceland has the largest production of Arctic charr in the world, but apart from Sweden, the species is also farmed in Canada, Norway and Great Britain. The majority of the strains used for food production are subject to selective breeding.

### 1.3 Sustainability

Sustainability has been defined as: “Development that meets the needs of the present without compromising the ability of future generations to meet their own needs” (Brundtland, 1987). During the 30 years that has passed since sustainability was defined, the concept has come to symbolise a concern for social, economic and environmental development, the three ground pillars of sustainability.

The concept sustainability is large, difficult to concretise and comprise many aspects (Olesen *et al.*, 2011). Since the concept involves all improvements striving for a safe environmental, social and economic development, full
assessments and hands-on benchmarks are either lacking or very diverse and inadequate (Vázquez-Rowe et al., 2012; Schau & Fet, 2008). Many factors can be difficult or almost impossible to measure despite various assessment tools that are available such as Life Cycle Assessment (see e.g. d'Orbcastel et al., 2009; Diana, 2009; Rebitzer et al., 2004; Rees, 1992).

1.4 Sustainability and Arctic charr farming

To date, there is no generally accepted definition of sustainable aquaculture (Olesen et al., 2011; Frankic & Hershner, 2003). Despite the lack of a definition, it is generally accepted that fully sustainable aquaculture should be environmentally acceptable, economically viable and socially equitable. The goal to reach a fully sustainable industry has though been claimed to be impossible due to the dependence on finite resources, its use of water and its waste generating regime (Diana, 2009).

The industry of farming salmonids has developed fast during the last 40 years and farming methods have improved, which has increased sustainability and reduced the environmental impacts in some aspects (Grottum & Beveridge, 2007).

Along the development of the Arctic charr industry, increased knowledge on behaviour, physiology, handling and requirements have contributed to improvements in fish rearing and welfare for Arctic charr (Brännäs et al., 2008; Jobling et al., 1998). Additionally, the industry has brought job opportunities to rural areas of Sweden. Fish farms have created spin off effects such as processing industry and infrastructure that benefit these areas. Also more farfetched effects, like increased tourism have arisen. Ice fishing nearby fish farms is a very popular activity that brings income from fishing licenses, camping grounds and local shops.

Arctic charr farming face many of the same challenges as all salmonid aquaculture production and there are sustainability issues in Arctic charr farming that needs to be developed and improved. These issues include everything from reducing the dependency of catches of wild fish for fish feed to optimal management of the farm and it products. In the present thesis the sustainability tasks mainly focus on methods that contribute to reduce environmental impacts and improve the economic viability of fish farms without measuring or evaluating sustainability per se. These factors are alternative protein sources, a shortened production cycle through selective breeding and the development of an adaptive feeding management model.
1.4.1 Feed ingredients

The commercial feeds used in fish farms for salmonids have traditionally contained fishmeal and fish oil, ingredients that fulfil most of the nutritional needs of farmed fish (Jobling 2001). Fish based raw materials have been questioned from a sustainability perspective since they potentially could be used as human food directly (Kiessling, 2009; Tacon & Metian, 2009; De Silva & Turchini, 2008), as prey fish for other marine species (Fréon et al., 2005), or because they come from poorly managed fisheries that cause damage to wild stocks (Deutsch et al., 2007). In 2015 only 12% of the global catch for fishmeal and oil came from stocks considered to be in “very good shape”. Most stocks, 62.5%, were reasonably well managed but the fisheries did not meet the standards for eco certifications. More than one third of all fish caught for inclusion in fish feed came from stocks that were poorly managed (Veiga et al., 2015).

Alternative protein sources

Changes of feed compositions where much of the fishmeal and oil has been replaced by plant derived proteins and oils have been made to decrease dependence on marine resources (Ytrestøyl et al., 2015; Skretting, 2013). The plant protein fraction in the pelleted fish feed has increased but the new ingredients such as soy, sometimes also face sustainability problems (Torrissen et al., 2011; Gatlin et al., 2007).

From a resource-efficient perspective, ingredients in animal feed can preferably be made up from materials that are not attractive for direct human consumption (Kiessling, 2009; Gatlin et al., 2007). Ingredients can also be derived from waste, such as trimmings (Chamberlain, 2011) or by-products from other industries (Øverland et al., 2013b; Tacon et al., 2006). Attention has also been brought to local or regional products that exist in smaller supplies for inclusions in fish feed for example algae, co-products from the nut industry and mussels (Arnason et al., 2015; Barrows & Frost, 2014).

There are many things to consider when introducing new ingredients in a fish feed. For example, suboptimal or too large inclusions of many plant derived proteins have been shown to negatively impact fish welfare (Geurden et al., 2009; Olsen et al., 2001), feed utilisation (Krogdahl et al., 2003), nutritional content of the fish flesh (Bell et al., 2001), digestibility and the levels of waste discharge (Schneider et al., 2004). Alternative protein sources that are not plant based such as invertebrates (Kroekel et al., 2012) and single cell proteins (Øverland et al., 2013a; Oliva-Teles & Gonçalves, 2001) has also been experimentally tested in fish feed with varying success. Another aspect
important to consider when introducing new ingredients is acceptability. Since fish in a farming environment always are subjected to only one type of food, it is important that they find it appealing (Raubenheimer et al., 2012). Many fish species show taste preferences (Kasumyan & Döving, 2003) and since feed ingredients affect not only the nutritional value but also the taste, the acceptability of the pellets may be influenced (da Silva et al., 2015). Reduced acceptance for a feed will have effects on the welfare of fish, increase the environmental load and reduce the economic viability due to lower growth (Jobling, 2001).

1.4.2 Selective breeding
Selective breeding for faster growth has been the main aim for all European fish breeding programs (Janssen et al., 2016). A faster growth shortens production time and improves the economic viability of the business (Eriksson et al., 2010). Breeding can also contribute to a more efficient farming through a more uniform growth that reduce the need for sorting and reduced early sexual maturation (Gjedrem, 2012; Gjedrem, 2010). Another effect of faster growth has been better feed utilization that decrease waste and improve utilization of resources (Gjedrem, 2005; Thodesen et al., 1999).

As the industry develops, breeding targets and methods for selection also evolve. Disease resistance has been a desirable trait that has been selected for during a long time but also processing yield and efficient feed utilisation occur in some breeding programs (Janssen et al., 2016; Gjedrem & Robinson, 2014). In addition, as new feed compositions and ingredients emerge, a desire to select for best possible growth or feed utilisation on new alternative feeds has been expressed (Quinton et al., 2007a; Quinton et al., 2007b; Kause et al., 2006).

1.4.3 Feeding management
Fish feed constitutes 50-70 % of the costs in intensive aquaculture (FDIR, 2010; Kiessling, 2009), why the development of accurate feeding schedules are essential for a more sustainable farming. Good feeding management is to feed the fish when they are hungry and to distribute the feed as equally as possible between all individuals (Alanärä et al., 2001). Key factors are the size of daily rations, frequency of feed portions and timing and distribution of meals, which depends on both biotic and abiotic factors (Goddard, 1995).

Feeding management in Swedish Arctic charr farms rely mainly on feeding charts and models supplied by the feed companies and feed system companies. These are typically based on water temperature and body size, but give little
possibilities for the farmer to make adjustments based on local conditions and the particular fish stock being held. This may lead to feed waste that have negative impacts on the environment and the economy of the farm (Smith et al., 1993). Alanärä et al. (2001) presented an adaptive model to calculate feed rations based on the daily energetic requirements of fish. The model is flexible and give room for local adjustments. The model has so far been tested with promising results on rainbow trout (Bailey & Alanärä, 2001), Atlantic salmon (Salmo salar) smolt (Alanärä et al., 2014), and Eurasian perch (Perca fluviatilis) (Alanärä & Strand, 2015).

Arctic charr show a strong seasonality, meaning that their appetite and growth capacity vary depending on season (Damsgard et al., 1999; Saether et al., 1996; Tveiten et al., 1996; Pálsson et al., 1992). They typically peak in growth and appetite in early summer to display low or no appetite and growth in late autumn. From a fish farming perspective this seasonality makes it difficult to adjust daily feed rations during different parts of the year. Thus, an adaptive feeding management model must include the possibility to handle seasonal variation in appetite.

1.5 Aims of the thesis

This thesis aims to explore the topics alternative protein sources in fish feed, a shortened production cycle through breeding, and feeding management. Factors that in their implementation may contribute to a more sustainable farming of Arctic charr. Specific objectives were to:

- Evaluate a new feed containing protein sources that were of low or no interest for human consumption through a preference experiment (paper I), a growth trial (paper II) and a sensory evaluation (paper II).
- Investigate possible family differences in growth response between the new alternative feed and a control feed (paper III).
- Investigate the effects of selective breeding on growth and seasonal growth capacity (paper IV).
- Apply new knowledge regarding growth capacity and seasonality and combine it with a new model for the energy need of Arctic charr to make an adaptive feed budget (paper IV).
2 Materials and Methods

2.1 Experimental design, fish and rearing

In all experiments, fish from the Swedish breeding program for Arctic charr, Arctic superior (Nilsson et al., 2010) were used. All fish were hatched at Aquaculture Centre North (ACN) in Kålarne, Sweden. Before handling, fish were always anaesthetized to reduce stress using MS222 (tricaine methanesulfonate, 40 mg l⁻¹) and very few mortalities occurred during experiments. All experiments were approved by the ethical committee of Northern Sweden, Umeå. Paper I: A13-13, paper II and III: A62-10.

2.1.1 Paper I

The trial was carried out at the Umeå Marine Science Centre (UMSC) in Norrbyn to investigate the preference of experimental feeds through self-selection. Sixteen Arctic charr were placed in aquaria divided into two compartments with a passage in the middle that allowed the fish to swim freely between the two compartments. Feeders were placed above each of the two compartments. Feed was given in excess twice daily: The test feed was distributed into one compartment of the aquaria, and the control feed in the other. Uneaten pellets were collected from each tank daily with a small net and thereafter counted. After nine days, the fish exhibited a clear preference for one of the feeds, and the positions of the feeds were reversed in all aquaria to control for left-right biases. The trial thereafter continued for another seven days, resulting in a total trial time of 16 days.
2.1.2 Paper II and III

In paper II, the effects of a new feed composition on growth and digestibility were investigated (feeds are described in detail in section 2.2) and the final fillets were evaluated in a sensory evaluation. In paper III, the growth response of ten full-sib families’ towards the experimental feeds were evaluated.

For paper II and III, data were obtained from an experiment conducted at ACN. In October 2012, 2970 juvenile fish were individually tagged with PIT-tags (Passive integrated Transponders, Biomark HPT12). The fish were divided into six groups of 495 fish each. Each of the six tanks initially held 33 fish from 15 different families. Fish had an average initial mass of 32.7 ± S.D. 10.1 g.

The feeding trial began in February 2013 and ended in December the same year. Temperature ranged from 1.2 to 13.8 °C (± 0.1 °C) during the experiment. Fish weight, fork length and general condition were recorded in February, May, September and December the same year for the evaluations.

In May and September, a thinning of the fish was conducted to ensure suitable biomass in the tanks and the welfare of the fish. In May, the 195 smallest individuals from each tank, 13 from each family were removed from the experiment. In the September thinning, five full families were removed from the tanks. The families that were excluded were: one of the best growing, one of the worst growing and three intermediately growing families. In December 2013 the trial ended, final weights and lengths were measured.

For paper II, digest was collected from the distal intestine (from the ileo-rectal valve to the anus) from eight randomly selected fish from each tank in September and December via dissection. The samples were pooled in one bulk sample per tank (three controls and three test feed) and stored at -20 °C until analysed. Muscle was sampled as a Norwegian cut on ten fish from each treatment to analyse total fat content and fillet colour at termination. In addition, 20 fish from each feed treatment were subject to standard slaughter procedures. Thereafter, the intestines were removed; fish were rinsed and stored on ice until prepared for a sensory evaluation.

2.1.3 Paper IV

An evaluation of the Swedish Arctic charr breeding programs’ effect on growth and seasonal variation in growth capacity was conducted. Data was obtained from the breeding program and different scientific trials. Three studies using fish from the first breeding generation and three studies from the sixth and seventh breeding generation. All data was collected at ACN and with exception for the water temperature regime for eggs and fry, all fish were been subjected to a similar rearing environment. Fish from the first generation were reared in
ambient water temperatures. In mid-May the start feeding began and the fry reached one gram by mid-July. During the second generation (in 1990) the facilities were improved so that the water for eggs and fry could be heated to 6 °C, resulting in an earlier hatching and start-feeding. The water was thereafter kept at 6 °C for fry until the natural temperatures reached 6 °C in May. Start feeding then began in late March and by June, fry reached one gram. For all generations the fry were reared in plastic tanks (0.8 m³). As they reached 30-50 g they were transferred to large concrete pools and kept under ambient water temperatures. Feeding was conducted to satiation.

To compare occurrence of early sexual maturation between generations, three datasets from the first generation and three from the sixth and seventh generation were used.

To make a model for the digestible energy need (DEN) to produce one unit of weight gain for Arctic charr, data came from trials with exact feed collection performed both at ACN and UMSC. The majority of the studies were conducted within the frames of this PhD-project and all are previously unpublished experiments. Feed intake and growth on individual fish was recorded from a lab set up (described in Strand et al., 2007) while fish on group level were recorded in a farm-like setting at ACN with feed waste collection (Hølland Teknologi, Norway).

2.2 Experimental feeds

We composed a new fish feed for Arctic charr called the “Baltic Blend” containing protein sources that are unattractive for human consumption (Kiessling, 2013; Eriksson et al., 2010; Kiessling, 2009). The protein sources were Baltic Sea fishmeal from two pelagic fish, sprat (Sprattus sprattus) and herring (Clupea harengus), Baltic Sea blue mussels (Mytilus edulis) and baker’s yeast (Saccharomyces cerevisiae) and the feed is evaluated in this thesis.

In paper I-III the same iso-nitrogenic and iso-energetic feeds were used, the Baltic Blend test feed and a fishmeal-based control feed mirroring a commercial type Arctic charr feed (Table 1). The Finnish Game and Fisheries Research Institute manufactured the experimental feeds at the Laukaa Aquaculture station in Finland. To enable traceability in the digestibility evaluations, titanium dioxide (TiO₂) was added to both diets.
The mussel meal in the test feed originated from the southwest region of the Baltic Sea and was obtained from Royal Frysk Muscheln GmbH Emmelsbüll-Hornsbül, Germany. The fishmeal in the test feed was decontaminated and came from the Baltic Sea (Triplenine, Denmark), and the fishmeal in the control feed originated from the Atlantic Ocean (Raisioagro Ltd., Raisio, Finland). The baker’s yeast was cultured on molasses, ammonia, phosphorus, magnesium and vitamins and then dried on a fluidized bed (Jästbolaget®, Stockholm, Sweden). The oil components of both feeds contained commercial fish oil and regionally produced rapeseed oil (Raisioagro Ltd., Raisio, Finland). The feeds chemical composition, energy content and amino acid content are presented in Table 2.

2.3 Chemical analyses

2.3.1 Betaine content, paper I
The amounts of betaine (glycine-betaine) in the feeds were analysed in a Bruker 600 MHz 1H NMR spectrometer with a zgessgp pulse sequence (Bruker Spectrospin Ltd., BioSpin, Karlsruhe, Germany).
Table 2. Chemical composition (g kg⁻¹ DM), energy content (MJ kg⁻¹ DM), betaine content (μ mol g⁻¹), and amino acid content (g kg⁻¹ DM) of the control and test (Baltic Blend) diet for Arctic charr.

<table>
<thead>
<tr>
<th></th>
<th>Control diet</th>
<th>Test diet</th>
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<tbody>
<tr>
<td>Crude protein</td>
<td>467</td>
<td>474</td>
</tr>
<tr>
<td>Sum of amino acids</td>
<td>457</td>
<td>408</td>
</tr>
<tr>
<td>Crude lipid</td>
<td>187</td>
<td>194</td>
</tr>
<tr>
<td>Ash</td>
<td>68</td>
<td>78</td>
</tr>
<tr>
<td>Gross energy</td>
<td>22.9</td>
<td>23.0</td>
</tr>
<tr>
<td>Betaine</td>
<td>5.4</td>
<td>22.7</td>
</tr>
<tr>
<td><strong>Indispensable amino acids</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Arginine</td>
<td>29.8</td>
<td>24.7</td>
</tr>
<tr>
<td>Histidine</td>
<td>10.6</td>
<td>8.8</td>
</tr>
<tr>
<td>Isoleucine</td>
<td>21.1</td>
<td>17.9</td>
</tr>
<tr>
<td>Leucine</td>
<td>35.9</td>
<td>29.1</td>
</tr>
<tr>
<td>Lysine</td>
<td>31.3</td>
<td>29.0</td>
</tr>
<tr>
<td>Methionine²</td>
<td>14.6</td>
<td>14.2</td>
</tr>
<tr>
<td>Phenylalanine</td>
<td>20.9</td>
<td>17.6</td>
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<tr>
<td>Threonine</td>
<td>18.2</td>
<td>17.2</td>
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<tr>
<td>Valine</td>
<td>25.2</td>
<td>21.7</td>
</tr>
<tr>
<td><strong>Sum</strong></td>
<td>207.4</td>
<td>180.2</td>
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<tr>
<td><strong>Dispensable amino acids</strong></td>
<td></td>
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<tr>
<td>Alanine</td>
<td>24.5</td>
<td>21.5</td>
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<tr>
<td>Aspartic acid</td>
<td>44.5</td>
<td>37.8</td>
</tr>
<tr>
<td>Cysteine³, ⁴</td>
<td>12.2</td>
<td>14.2</td>
</tr>
<tr>
<td>Glutamic acid</td>
<td>79.1</td>
<td>67.8</td>
</tr>
<tr>
<td>Glycine</td>
<td>24.2</td>
<td>23.8</td>
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<tr>
<td>Ornithine</td>
<td>2.0</td>
<td>6.2</td>
</tr>
<tr>
<td>Proline</td>
<td>23.7</td>
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</tr>
<tr>
<td>Serine</td>
<td>21.6</td>
<td>18.8</td>
</tr>
<tr>
<td>Tyrosine⁴</td>
<td>17.7</td>
<td>16.4</td>
</tr>
<tr>
<td><strong>Sum</strong></td>
<td>249.4</td>
<td>228.2</td>
</tr>
</tbody>
</table>

¹ Dry matter values for both diets were 97 %
² Amount present after oxidation of methionine to methionine sulphone.
³ Amount present after oxidation of cysteine and cystine to cysteic acid.
⁴ Conditionally indispensable (NRC, 2011).

2.3.2 Feeds and faeces paper I, II and III

Faeces and experimental feed were freeze-dried, grinded with a coffee grinder (KG40, DeLonghi Appliances, Italy) and stored at -25 °C until analyses. The ash content of each feed was determined after samples were incinerated at 550 °C for a minimum of three hours. Dry matter (DM) was determined after heating
the samples in an oven in 103 °C for 16 h and then cooling them in desiccator before weighing. Total nitrogen (N) was determined by the Kjeldahl method using a 2020 digester and a 2400 Kjeltec Analyser unit (FOSS Analytical A/S, Hillerød, Denmark). The Crude protein (CP) content was determined through total nitrogen (N) using the Kjeldahl method calculated as N × 6.25 (Nordic Committee on Feed Analysis, 1976). Crude lipid (CL) levels were determined according to the Official Journal of the European Communities (1984) using a hydrolyzation and extraction system (1047 Hydrolysing Unit and a Soxtec System HT 1043 Extraction Unit, FOSS Analytical A/S, Hillerød, Denmark). Gross energy (GE, MJ kg⁻¹) of the feeds was determined using an isoperobol bomb calorimeter (Parr 6300, Parr Instrument Company, Moline, IL, USA). Titanium dioxide was analysed according to Short, et al. (1996).

Amino acid content in the feed and faeces was determined as described by Abro Rani et al. (2014) using the AccQ·Tag™ method (Waters Corporation, Milford, MA, USA). The samples were hydrolysed in 15 ml 6 M HCl containing 1 % phenol in a microwave oven (Synthos 3000, Anton Paar Nordic, AB Sweden). For analyses of cysteine and methionine, 50 mg feed samples were added to 2 ml formic acid: perhydrol (9:1) and incubated overnight at +4 °C. Thereafter, 2 ml of a freshly prepared sodium bisulphite solution (0.17 g ml⁻¹) were added and samples were mixed for 15 min. The samples were then hydrolysed and thereafter neutralised, diluted and derivatised according to the Waters UPLC® amino acid analysis solution protocol. The UPLC system was a Dionex, Ultimate 3000 binary rapid separation LC system with a variable UV-detector (Thermo Fisher, Sweden, Stockholm). Empower 2 (Waters) software was used for system control and data acquisition.

2.3.3 Fat content, paper II
The muscle fat content was analysed from the fillet samples. Approximately 3 g from the left fillet were homogenised in a food processor together with an alkaline detergent (LOSsolvér Fish, MIRIS AB, Uppsala, Sweden) at 45 °C. Thereafter analysed using a Mid-Infrared-Transmission (MIT) spectrooscope (MIRIS AB, Uppsala, Sweden) according to the manufacturer’s instructions. The average MIT value of two to three sub-samples of each homogenised fillet was used.

2.3.4 Flesh colour, paper II
Carotenoid (astaxanthin) levels in the fillet samples were analysed using a simplified method for total carotenoid content (Torrissen, 1986). Astaxanthin
and other carotenoids in the muscle were extracted in acetone followed by evaporation and dilution with isopropanol. Total carotenoids were measured by spectrophotometry at 477 nm.

### 2.4 Microscopy, paper II

The faeces were investigated with microscopy. For vital staining of the fish faeces samples a LIVE/DEAD® BacLight™ Bacterial Viability Kit (Molecular Probes, Eugene, OR, USA) was used. The samples of frozen faeces were diluted in 150 μL of 0.85 % NaCl and 50 μL of dye solution (10 μL Component A + 10 μL Component B + 980 μL 0.085 % NaCl), vortexed and thereafter incubated. One droplet was put on a glass slide and covered with a coverslip. To observe yeast cells in the samples a microscope coupled to a HGFI mercury lamp and a camera (Nikon, Tokyo, Japan) was used. Bright field and epifluorescence images were obtained. Green (Epi-FL Filterset FITC, excitation wavelength 470-490 nm, emission 520-560 nm) and red (Epi-FL Filterset Texas Red, excitation 540-580 nm, emission 600-660 nm) light fluorescence filters were used to observe the SYTO 9 and propidium iodine fluorescence, respectively.

### 2.5 Sensory evaluation, paper II

Fish were filleted and the filets were cut into three parts. The pieces were cooked in a steam oven for 4 min and 58 sec at 52 - 54 °C (Jonsson et al., 2007). A panel with 26 members, staff and students at Umeå University School of Restaurants and Culinary Arts performed the sensory evaluation. The intensity of the sensory attributes flavour, odour, texture and appearance were scored according to the profiling method, Generic Descriptive Analysis (Stone & Sidel, 1985) and was performed according to Arnason et al. (2013) with exception for panel member training. Participants were asked to grade the intensity of five to seven adjectives for each sensory attribute on a scale of 0-100. The evaluation was a blind-test.

### 2.6 Calculations and statistical methods

Growth was measured using different methods, in paper I the specific growth rate (SGR) was calculated:

\[
SGR = \frac{(\ln W_2 - \ln W_1) \cdot 100}{t}
\]
where $W_1$ is the initial weight (g), $W_2$ is the final weight (g), and $t$ is the number of days between $W_1$ and $W_2$. In paper II, III and IV growth capacity was calculated as thermal growth coefficient, TGC (Cho, 1990) expressed as:

$$TGC = \left(\frac{W_2^{1/3} - W_1^{1/3}}{T \cdot D}\right) \cdot 1000$$

where $W_1$ is initial weight and $W_2$ final weight, $T$ is water temperature ($^\circ$C) and $D$ the number of days between $W_1$ and $W_2$. In paper II, Fulton’s condition factor (K) was calculated, expressed as:

$$K = 100 \cdot \left(\frac{W}{L^3}\right)$$

where $W$ is weight (g) and $L$ is fork length (cm).

An additional growth index was calculated in paper III to neutralize the effect that feed treatment and seasonal growth capacity had on growth. Each individual fish TGC was expressed in relation to the average TGC within each tank and period. Average values were thereafter composed per family and tank.

Economic feed conversion ratio, eFCR was calculated as feed fed (g)/biomass gain (g) in paper II.

The apparent digestibility coefficient, ADC was calculated in paper II for dry matter, crude protein, lipid, energy and indispensable amino acids, respectively. ADC was calculated according to Cho et al. (1982) as:

$$ADC = 1 - \left(\frac{F}{D} \cdot \frac{D_i}{F_i}\right) \cdot 100$$

where $F$ is % nutrient content of faeces, $D$ is % nutrient content of diet, $D_i$ is % digestion indicator of diet (TiO2), and $F_i$ is % digestion indicator of faeces.

The digestible energy need, DEN (kJ DE g⁻¹) describes the amount of energy (kJ DE) the fish needs to ingest to increase 1 g in wet weight and was calculated in paper IV as:

$$DEN = \frac{FI \cdot DE}{(W_2 - W_1)}$$

where $FI$ is the feed intake (g), $DE$ is the digestible energy content of the feed (kJ g⁻¹), $W_1$ is the initial weight (g) and $W_2$ the final weight (g) of the fish.

The theoretical daily energy requirement, TER (kJ day⁻¹) (Alanärä et al., 2001) calculated in paper IV was expressed as:

$$TER = TW_i \cdot DEN$$

where $TW_i$ is the theoretical daily growth increment (g day⁻¹) and DEN the digestible energy need (kJ DE g⁻¹).
2.6.1 Growth simulations, paper IV

To evaluate growth improvements within the breeding program growth simulations were conducted.

To account for variation in growth capacity at different times of the year, data from three out of the six studies in the total comparison between generations was used to create a seasonal growth capacity factor. The fish in these studies were weighed at monthly intervals over the whole study period. The growth capacity factor was calculated by dividing all monthly values on TGC by the highest value in each study (obtained in March). The relationship between time of year (Julian day, JD) and growth capacity factor (GCF) was described as; $GCF = 6.04E^{-8} \cdot JD^3 - 4.13E^{-5} \cdot JD^2 + 6.40E^{-3} \cdot JD + 0.494$ (Figure 1).

![Figure 1](image-url)

*Figure 1. Seasonal growth profile of Arctic charr, adjusted values (Growth capacity factor). Circles denotes values from 1985-86, triangles 1986-87 and squares values from 2012-13.*

The growth capacity factor was thereafter multiplied with the TGC value when solving for $W_2$ in the TGC equation, enabling calculations of the daily growth ($W_2$). This equation was:

$$W_2 = \left(W_1^{\frac{1}{3}} + \left(\frac{TGC \cdot GCF}{1000} \cdot T \cdot D\right)^3\right)$$

To get the daily TGC values that equalled the theoretical weight with the real weight in all data sets the problem solver in Excel was used. It fills in the theoretical values on daily weights between weighing’s. This procedure created data that were comparable for both daily weight increment and TGC for all studies, despite differences in sampling intervals and dates.
2.6.2 Statistical methods

The level of significance was 0.05 for all statistical analyses.

In paper I, the preference of feeds was analysed with a repeated measures ANOVA, with feed intake as dependent variable, and feed as factor. To verify that pellets were not dissolved instead of eaten between collection occasions, linear regressions were made for test and control feed eaten as well as for total feed intake with the growth rate. Analyses were conducted using SPSS (IBM SPSS Statistics, Armonk, NY).

For paper II, possible differences between the two feed treatments in weight increase, TGC and K-factor were analysed using mixed model ANOVA in JMP® Pro 12 (SAS institute Inc. Cary, NC). Feed treatment was used as fixed factor and tank as random factor nested within each feed treatment. EFCR between the two treatments for the different time periods and differences in ADC of indispensable amino acids between the treatments in December were analysed using two-sample t-tests using the MINITAB® statistical software package (Version 16; Minitab, State College, Pennsylvania) and corrected for multiple comparisons with a false discovery rate (Benjamini & Hochberg, 1995). The effects of feed and sampling occasion on ADC were analysed, as was the interaction between these two factors using two-way ANOVA in SPSS 21.0. Sensory evaluation data were analysed using the Mann-Whitney u-test in MINITAB and corrected for multiple comparisons with a false discovery rate (Benjamini & Hochberg, 1995).

In paper III, families start weights were analysed with one way ANOVA using average family weight as response variable and family as factor. Any differences between tanks were investigated with one way ANOVA using families as response variable and tank as factor. Families’ responses to feed treatment and season were evaluated using the average TGC values for each family and tank during three periods (spring, summer and autumn). A mixed model ANOVA with a repeated measures structure and a full factorial design with family, feed and season was used. As a continuous factor average day number for each time period was included. Family averages were included as random factor nested per tank. A mixed model ANOVA with a repeated measures structure and a full factorial design was similarly used to investigate the effect of family and season on growth index. Family averages were included as random factor nested within each tank. All analyses were made using JMP pro 12.0.

In paper IV, the growth capacity differences between the generations were analysed with ANOVA, with TGC, temperature sum and daily weight gain as response variables, and generation and season as factors. The response variables were made up by a mean value of the three months in the three periods winter
(February, March and April), summer (June, July and August) and autumn (October, November and December) respectively. Effects of body weight and temperature on DEN, were analysed with ANOVA. The analyses were made using JMP pro 12.
3 Main results

3.1 Paper I

The behavioural approach self-selection can successfully be used for single reared Arctic charr in aquaria to investigate acceptance of new diets. Arctic charr ingested more test feed than control feed in the experiment ($F_{15,420}=1.53$, $p=0.046$, Figure 2).

Figure 2. Daily feed ratio (eaten test feed/control feed). A value larger than one shows that fish ingested more pellets of the test feed and a value lower than one that fish ate more control feed. Error bars indicate S.E.M. The black dashed line indicates the time at which the feeds were reversed in the aquaria.
3.2 Paper II

The overall growth response of the fish fed the Baltic Blend test feed was 11.5 % lower than fish fed a control feed, F=26.19, p=0.007, Table 3). Digestibility of the test feed was lower than for the control feed for dry matter, crude protein, lipid and gross energy (Table 3). A microscopy analysis revealed whole yeast cells that had not been digested in the faeces of fish fed the test feed. Additionally, analyses of the test feed revealed a lower amino acid content compared with the control feed.

Table 3. Final weights (g) of fish fed the test and control feed respectively. Apparent digestibility coefficient (%) for dry matter, crude protein, lipid, and gross energy of the Baltic Blend test feed and the control feed for Arctic char, (± S.E.M.), n=3, Digestibility values only from the end of the growth trial displayed (December 2013).

<table>
<thead>
<tr>
<th></th>
<th>Test</th>
<th>Control</th>
<th>t</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Final weight (g)</td>
<td>591.2 (13.8)</td>
<td>667.5 (5.2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Digestibility</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dry matter</td>
<td>67.4 (0.94)</td>
<td>74.6 (1.15)</td>
<td>4.87</td>
<td>0.017</td>
</tr>
<tr>
<td>Crude protein</td>
<td>77.4 (0.33)</td>
<td>88.9 (0.37)</td>
<td>23.1</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>Lipid</td>
<td>83.2 (2.22)</td>
<td>93.4 (0.48)</td>
<td>4.48</td>
<td>0.046</td>
</tr>
<tr>
<td>Gross energy</td>
<td>73.2 (0.72)</td>
<td>81.4 (0.72)</td>
<td>7.97</td>
<td>0.004</td>
</tr>
</tbody>
</table>

In the sensory evaluation of the filets a panel of 26 participants rated attributes within the categories flavour, texture, odour and appearance similar for the two feed treatments (Figure 3).
3.3 Paper III

An evaluation of full-sib family differences in growth response towards the Baltic Blend feed in comparison to a control feed was conducted. Fish fed the control feed grew better than fish fed the test feed. No effect of the interaction between feed and family was found and the families that grew well on the control feed were also the families that grew well on the test feed (Figure 4). The families displayed varying growth and also had a divergent seasonal growth pattern where some families grew better in summer and others during autumn (Figure 4 and 5). The interaction for feed, family and season was not significant.
Figure 4. Growth capacity (TGC) of the fish fed the Baltic Blend test feed and the control feed for the ten families of Arctic charr during the experiment (February to December). Error bars denote S.D.

Figure 5. A post-hoc test showing the interaction between season and family on growth index (Overall average decision chart, JMP® Pro 12). The horizontal lines show 95% confidence interval. Families that diverge in growth in relation to the average growth within each tank display values outside the confidence interval.

3.4 Paper IV

Our investigations of growth improvements and seasonal growth capacity in Arctic charr as an effect of the Swedish breeding program revealed that the
heating of water to 6 °C during egg incubation and the fry stage has affected growth and hatching time for Arctic charr. The heating alone may have resulted in slightly more than 20 g weight improvement by December after the first growing season (Figure 6).

Figure 6. Weight increase of Arctic charr in their first growing season (hatching to mid December). Circles denote weight measurements from the first generation and squares weight measurements from the present generation. The dotted line shows the growth pattern for the first generation in ambient temperatures, the hatched line is a simulation of weight development of fish from the first generation adjusted for heating of water for eggs and fry to 6 °C.

A growth simulation based upon seasonal variations in TGC was constructed and illustrate the improvements in growth capacity between the first and present generations of Arctic charr (Figure 7). Comparisons between generations during the second year of rearing show that TGC has improved with generation (ANOVA, F1,16=11.74, p= 0.005). TGC has improved 1.8 times in winter (Feb, Mar and Apr), 1.4 times in summer (Jun, Jul and Aug) and 1.2 times in autumn (Oct, Nov and Dec).
Breeding of Arctic charr has resulted in a faster growing fish, reaching slaughter size of 657 g (S.D ±17), ten months faster than for fish from the first generation in the breeding program when adjusted for improved rearing conditions (Figure 8).

**Figure 7.** TGC profile for Arctic charr from generation one (filled circles) in the breeding program and generation six and seven respectively (empty circles) from about five months age. Error bars denote S.D.

**Figure 8.** Simulated weight development for Arctic charr from generation six and seven (whole line) and generation one (hatched line) in the breeding program. Gray fields symbolize S.D. Squares denote actual weight measurements from generation six and seven and circles from the first generation. The dotted line denotes a growth simulation of the first generation adjusted for heating of water for eggs and fry.
The daily weight increase was different between generations (ANOVA, $F_{1,16} = 41.13$, $p<0.001$), seasons (ANOVA, $F_{2,16} = 59.02$, $p<0.001$) and the interaction between generation and season was significant (ANOVA, $F_{2,16} = 8.45$, $p=0.005$). The interaction display that the differences in the daily weight increase was the highest between the generations in summer followed by autumn and thereafter winter. The total improvement in weight was 11 % per generation when adjusted for heating of water for eggs and fry.

A new model for the digestible energy need of Arctic charr was created for fish in the size range of 20-850 g. It was constructed by curve estimation, and a linear fit explained the data best; $DEN = 9.7099 + 1.246 \times \ln (BW)$, $(R^2=0.47)$ (Figure 9).

Figure 9. The relationship between body weight and digestible energy need (DEN, kJ DE g$^{-1}$) of Arctic charr. The black circles symbolize fish on group level and the gray circles single reared fish. The dotted line is the relationship for the DEN-model. Error bars denote S.D.
4 Discussion

The examples in this thesis may contribute to increase sustainability of Arctic charr farming. The factors that have been in focus; alternative protein sources, a shortened production cycle through selective breeding and an adaptive feeding management model would need sustainability assessments that comprise all aspects, environmental, economic, and social, to make solid predictions regarding their effects on sustainability of Arctic charr farming. Below, I discuss the results and their potential contribution to a more sustainable farming of Arctic charr.

4.1 Alternative protein sources

When searching for new ingredients in fish feed, alternatives to potential human food sources is a favourable approach (Kiessling, 2009; Tacon & Metian, 2009; Gatlin et al., 2007). The protein fraction in the Baltic Blend feed evaluated in this thesis (paper I, II and III) contained Baltic Sea blue mussels and decontaminated fatty fish (sprat and herring) from the Baltic Sea and additionally baker’s yeast. All three ingredients are unattractive for direct human consumption and have promising properties for inclusion in feed for Arctic charr.

The sprat and herring are two fatty fish species that in the Baltic Sea contain high levels of Polychlorinated biphenyl (PCB) and dioxins, making them unattractive for human consumption (Isosaari et al., 2006). An industrial process can separate the toxic compounds from the meat, which enables inclusion in fish feed (Cheng et al., 2016; Sprague et al., 2010; Oterhals & Nygård, 2008). Even if the cleaning process is efficient, evaluations are lacking of the potential accumulation of harmful substances in Arctic charr from the Baltic Sea fishmeal.

Blue mussel meal from the Baltic Sea has a favourable amino acid profile for Arctic charr (Langeland et al., 2016), as well as a fat composition suitable for
salmonids (Arnason et al., 2015; Berge & Austreng, 1989). Recent research show a good growth response in rainbow trout when fed a feed with blue mussel as main protein ingredient (Arnason et al., 2015). The mussels are good nutrient assimilators, but due to a low salinity in the Baltic Sea, they never reach full market size (Schütz, 1964) and are therefore not attractive for human consumption. Mussels have also been suggested to increase palatability of feed for some fish species as they contain betaine (Kasumyan & Döving, 2003; Meyers, 1987; Mackie et al., 1980). Chemical analysis of the Baltic Blend feed showed four times higher levels of betaine in the test feed (paper I) and this might have acted as a feeding stimulant in the Baltic Blend feed (paper I).

Baker’s yeast has a potential to grow fast at a low cost. It may also grow on substrates such as waste biomass and thereby utilize resources more efficiently or even recycle nutrients back into the human food chain (Matassa et al., 2015; Gelinas & Barrette, 2007). High levels of nucleic acids in yeast limits the possibilities for its usage in feeds for terrestrial animals, but not for fish (Kinsella et al., 1985). Yeast in fish feed for salmonids has revealed some difficulties with digestibility (Øverland et al., 2013a; Rumsey et al., 1991; Rumsey et al., 1990). However, prior to the formulation of the Baltic Blend feed used in paper I, II and III, shorter experiments with Arctic charr were conducted showing that Arctic charr, in terms of growth and digestibility, responded well when fed feeds with intact yeast (Langeland et al., 2016; Vidakovic et al., 2015). These results motivated the inclusion of whole yeast cells in the Baltic Blend feed. The relatively long trial time of ten months most likely highlight and expand the effects that the lower digestibility found in paper II has on growth. The microscopy investigation of faeces in paper II most likely showed whole yeast cells that had not been utilized by the fish. Our results emphasize the benefits with conducting longer trials. Additionally, we found a lower amino acid content in the test feed that also may have influenced growth capacity negatively. We suspect that the yeast inclusion needs to be modified in some respect to increase digestibility and growth.

A new feed needs to be evaluated in many different ways. Low palatability of a feed may increase feed waste and hinder growth (Jobling, 2001), which has a large influence on both environmental and economic sustainability of the production. Evaluating the fish acceptability of new diet compositions is a way to improve the welfare of fish, sustainability and success of new diets (da Silva et al., 2015). Self-selection, the method used in paper I, works advisory for making a first prediction regarding a feeds suitability, an important first step before conducting large, expensive life- and time-consuming experiments (da Silva et al., 2015) such as the long-term study in paper II. The study in paper I is to my knowledge the first time a self-selection study on pelleted feeds with
different ingredients has been made with Arctic charr. The preference Arctic charr showed for the Baltic Blend test feed over the control feed (paper I) suggest that the lower growth obtained in paper II was not a result of feed rejection caused by poor palatability.

The taste and quality of the fish flesh may also be affected by a new feed composition (Lie, 2001) and the economic profit will be impaired if consumers are hesitant towards the final product. The Arctic charr produced on the Baltic Blend feed gained approval among consumers, similar opinions were expressed for the two treatments in our study (paper II).

It has been suggested that the ingredients in the Baltic Blend feed originating from the Baltic Sea can create a nutrient loop through uptake and transport of nutrients from the eutrophicated Baltic Sea to the oligotrophic hydropower reservoirs, where Arctic charr mainly are farmed (Eriksson et al., 2010; Kiessling, 2009). Hydropower reservoirs, with high water level amplitude are heavily disturbed ecosystems in which the littoral zone as a result is damaged by waves and ice. As a result, the reservoirs have become further oligotrophicated and primary production has decreased dramatically (Stockner et al., 2000). As a result, the growth of wild fish decrease (Milbrink et al., 2011) and biological diversity is also threatened (Karlsson et al., 2009; Persson et al., 2008). To stimulate primary production, and ultimately fish production by nutrient addition may mitigate this type of ecosystem damage (Milbrink et al., 2011; Stockner & Hyatt, 1984). Studies in Sweden have shown positive ecosystem effects after nutrient addition and within a few years wild fish have recovered to a similar size as before impoundment, without negative effects on water quality and other food web components (Milbrink et al., 2011; Milbrink et al., 2008; Rydin et al., 2008).

Mussels are filter feeders and blue mussel farms can function as cleaners of nutrient rich waters (Lindahl et al., 2005). Blue mussel pilot farms have successfully been set up in the Baltic Sea (Lindahl, 2012). Together with the fatty fish species sprat and herring, nutrients can be removed from the Baltic Sea and transformed into Arctic charr biomass. Inevitably, farming in open systems causes nutrient rich waste, a nutrient addition of nitrogen, phosphorus and carbon. Thus, the similar ecosystem effects that can be seen from nutrient additions has been hypothesised to be achieved in the regulated water bodies by nutrient addition via fish farming (Eriksson et al., 2010; Kiessling, 2009). Parts of the nutrient transport concept has been theoretically evaluated for farming of Rainbow trout with promising results (Vrede, 2014). However, potential long-term ecosystem effects from the nutrient input that aquaculture cause in these systems has not yet been investigated.
The future development for alternative protein sources and feeds for Arctic charr is likely a similar path as for other commercially farmed salmonids in terms of ingredients used. However, Arctic charr is a promising species when it comes to reduced dependency on fishmeal. Protein levels in feeds for Arctic charr larger than 90 g can possibly be reduced without any growth reductions if lipid levels are increased (Sigurgeirsson et al., 2009), results that are highly interesting for the future development of Arctic charr feeds and farming.

4.2 Selective breeding

About 80% of all intensive aquaculture in Europe is conducted on stocks subjected to selective breeding (Janssen et al., 2016). Most traits included in breeding programs adapt fish better to the farming environment and thereby increase efficiency and the economic profit.

We found a growth progress of 11% per generation in paper IV, which is comparable to growth improvements achieved in other breeding programs for salmonid species (Janssen et al., 2016; Gjedrem & Robinson, 2014). The shortened production cycle, a result of the faster growth and also the lower occurrence of early sexual maturation, are both results of domestication of Arctic charr. The shorter production cycle influences the economic sustainability greatly (paper IV).

In the future, new traits will be included in breeding programs (Janssen et al., 2016) and selection for an efficient growth or feed utilization on alternative feeds is not an unlikely path for the industry (Quinton et al., 2007a). But our (paper III) and others (Quinton et al., 2007b) results show, that breeding for high growth in general will likely premier selection on alternative feeds too, as long as only modest modifications are made in feed compositions.

However, breeding also has its downsides as fish become increasingly different from its wild conspecifics, problems with escapees arise and wild populations of fish may be affected (Glover et al., 2012; McGinnity et al., 2003).

In the evaluations of different families’ growth response towards the new Baltic Blend feed (paper III) we also found some differences in growth capacity (TGC) among families at different times of the year. Similar findings have been reported earlier for this strain (Nilsson et al., 2016; Nilsson, 1992). This varying seasonal growth capacity may be used to further increase growth and thus the profit and sustainability in Arctic charr farming if implemented as a selection criteria in the breeding program. Good winter growth is a desirable trait considering the long winters that this species experience. However, the actual weight improvement is much larger during summer and a selection for better summer growth capacity may lead to a larger weight improvement.
4.3 Feeding management

Not only do the farmer risk economic loss but also negative environmental impacts are at stake when it comes to poor feeding management. Both underand over feeding of fish is associated with consequences. An underestimation of feed rations may increase aggressive interactions among individuals and lead to fin damages and other injuries (Persson & Alanärä, 2014). Underfeeding will likely result not only in a total lower biomass but also a less uniform stock and welfare problems. To overestimate growth capacity or appetite will instead result in economic loss and feed waste (Goddard, 1995). A poor consideration to seasonality has also been shown to result in feed waste (Smith et al., 1993). Furthermore, the knowledge of long-term environmental effects of effluent waste from Arctic charr farming in northern Sweden is to a large degree lacking, which further motivates to carefully study the growth pattern of the fish (paper IV) and using an attentive feeding management as the one presented in paper IV.

Models to calculate feed rations are commonly based on either energy requirements (Cho, 1992) or growth rate estimates (Austreng et al., 1987) but do rarely allow for modifications to suit local conditions. An adaptive model to estimate the daily energy requirements and to calculate feed budgets was developed by Alanärä et al. (2001). This model is based upon two components: the daily growth increase and the amount of digestible energy needed to obtain one unit of biomass increase. Both these can be retrieved in a normal farming situation and be updated to suit the local requirements.

The daily growth capacity (TGC) for the present generation of selected Arctic charr, from March 15th to December 31st, is described in paper IV and can be expressed as: 

\[ TGC = 771.6 \cdot JD^{-1.123} \]

where JD is Julian day. The theoretical daily weight gain (TW) can then be estimated on any given day, and together with values on the digestible energy need (DEN) (paper IV), the theoretical energy requirement (TER) can be calculated (Alanärä et al., 2001). TER can thereafter be used to calculate the daily feed allowance (FA) as;

\[ FA = \frac{TER \cdot n}{DE} \]

where n is the number of fish in the unit and DE is the digestible energy content of the feed (kJ g\(^{-1}\)). In table 4 and 5 an example is given on how the daily feed allowance of a group of Arctic charr can be calculated using this methodology.
Table 4. *Data and parameters for calculating the daily feed allowance.*

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Data</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fish size (g)</td>
<td>250</td>
</tr>
<tr>
<td>Temperature (°C)</td>
<td>12</td>
</tr>
<tr>
<td>Feed, digestible energy content (kJ g⁻¹)</td>
<td>19.0</td>
</tr>
<tr>
<td>Number of fish</td>
<td>5000</td>
</tr>
<tr>
<td>Date</td>
<td>July 5th</td>
</tr>
<tr>
<td>Julian day, JD</td>
<td>187</td>
</tr>
</tbody>
</table>

Table 5. *Example on how the daily feed allowance can be calculated for a group of Arctic charr.*

<table>
<thead>
<tr>
<th>Model</th>
<th>Calculation</th>
<th>Result</th>
</tr>
</thead>
<tbody>
<tr>
<td>TGC = 771.6 · JD⁻¹⁻¹²³</td>
<td>TGC = 771.6 · 187⁻¹⁻¹²³</td>
<td>2.17</td>
</tr>
<tr>
<td>W₂ = (W₁⁻¹/³ + (TGC / 1000 · T · D))³</td>
<td>W₂ = (250⁻¹/³ + (2.17 / 1000 · 12 · 1))³</td>
<td>253.1</td>
</tr>
<tr>
<td>TWᵢ = W₂ - W₁</td>
<td>TWᵢ = 253.1 - 250</td>
<td>3.1</td>
</tr>
<tr>
<td>DEN = 9.7099 + 1.246 · ln(BW)</td>
<td>DEN = 9.7099 + 1.246 · ln(250)</td>
<td>16.6</td>
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Half of all fish consumed by man is produced in fish farms. Aquaculture is a diverse industry that comprises many fish species and methods for farming. The industry has grown rapidly and improved markedly, however, there are still sustainability challenges to overcome.

In Sweden, aquaculture is a small industry, making up for 0.05% of the total global production. Mainly rainbow trout and Arctic charr are farmed and they are often sold as rather expensive products.

Arctic charr is a fish species that is well adapted to cold water conditions and it has a good ability to grow well in low temperatures, making it suitable for the climate in northern Sweden. Selective breeding for fast growth and late sexual maturation in a breeding program has enabled a reliable production in Sweden, providing a continuous supply of fresh fish to the markets. As a result, the Arctic charr farming has grown, between 2004 and 2014 the production increased fivefold to almost 1700 tons.

To decrease the dependency of unsustainable fishmeal from global catches in fish feed, there is a need for other more sustainable feed ingredients. A possible way forward is to use materials unattractive for human consumption, or by-products from industrial processes, for example distiller’s dried grains, co-

Popular science summary

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products from the nut industry or fish trimmings, to allow for a more efficient utilization of resources.

Information on how much feed a group of Arctic charr need in Swedish farms is often provided by the feed companies, figures that are difficult for the fish farmer to adjust to local conditions. It is especially hard to take into account seasonal variations in appetite and growth, which seldom are included in the feed companies’ recommendations. Breeding progress may also change the growth pattern of fish. To minimize feed waste that leads to economic loss and negative environmental impacts, an adaptive feeding management is a possible way forward to reach a more sustainable farming regime.

This doctoral thesis explores different actions that could improve the sustainability of Arctic charr farming such as alternative feed ingredients, a shortened production cycle through selective breeding and an adaptive feeding management model.

A new feed composition, the Baltic Blend, containing a protein mixture of decontaminated Baltic Sea fishmeal, Baltic Sea blue mussels and baker’s yeast was evaluated for Arctic charr in this thesis. Furthermore, the effects 30 years of breeding has had on growth, production time, and the seasonal growth pattern of Arctic charr was evaluated through data compilations and growth simulations.

In the evaluations of the Baltic Blend feed, Arctic charr displayed a lower growth on the new feed. This was likely caused by a poorer ability to digest the new feed due to the cell properties of the baker’s yeast. However, the fish preferred the Baltic Blend over a commercial type feed in a palatability test and consumers did not make any distinctions between fish fed the new feed and a commercial type feed. The results are promising but further development, for example through recipe modifications to increase growth are needed for the Baltic Blend to become a viable alternative to the commercial fishmeal and soy-based feeds used today.

Evaluations of 30 years of breeding of Arctic charr showed that it has had large positive effects on growth and production. The production cycle of Arctic charr has been shortened with ten months. The seasonal growth pattern of Arctic charr has also been affected by breeding, the fish now has a better growth capacity during winter than before.

A new model that can be used when calculating the daily amount of feed for Arctic charr was developed. Together with the new information on for example seasonal growth pattern it can be used as an adaptive feeding management model with increased reliability for local strains. If used in a real farming situation feed waste can likely be reduced and economic and environmental sustainability may be improved.
Sammanfattning på svenska

Hälften av all fisk som konsumeras globalt är odlad och vattenbruket har under de senaste årtiondena växt kraftigt. Trots industrins utveckling och förbättringar i produktionen så återstår många utmaningar för att förbättra hållbarheten.

Svenskt vattenbruk är en relativt liten industri som årligen producerar 0,05 % av den globala produktionen. Främst odlas regnbågslax och röding och de saluförs till stor del som exklusiva produkter över disk.


För att minska vattenbrukets beroende av fiskmjöl som ofta kommer från ohållbara fisken och potentiellt skulle kunna användas som mat till människor direkt behövs andra, mer hållbara ingredienser i fiskfoder. Möjliga alternativ är att istället använda råvaror som inte lämpar sig som människoföda eller biprodukter från industriella processer, exempelvis fiskrens, restprodukter från nötindustrin eller etanolframställning, vilka bidrar till en bättre resursanvändning.

Denna avhandling undersöker metoder som kan bidra till en ökad hållbarhet av rödingodling såsom alternativa proteinkällor i fiskfoder, en förkortad produktionscykel genom avel och en flexibel utfodringsmodell.

Effekterna av en ny fodersammanställning, ”Baltic Blend”, innehållande proteinkällor som inte är attraktiva för direkt mänsklig konsumtion utvärderades för röding i denna avhandling. Fodret innehöll renat Östersjöfiskmjöl, blåmusselmjöl från Östersjön samt jäst och det testades mot ett kontrollfodret som liknade ett kommersiellt rödingfoder. Vidare utvärderades effekterna av 30 år av avel på rödingens tillväxt och säsongsmönster genom datasammanställningar och tillväxtsimuleringar.


Rödingavel under 30 år har haft positiva effekter på bland annat fiskens tillväxt. Från den första generationen 1985 till den nuvarande sjunde generationen ökade tillväxten och produktionstiden förkortades med tio månader. Aveln har dessutom påverkat säsongsmönstret i tillväxtkapacitet som röding har och fisken hade en bättre tillväxtkapacitet under vintern än tidigare.

En ny modell som kan användas för att beräkna rödingens dagliga foderbehov utvecklades. Tillsammans med ny information rörande rödingens säsongsmönster i tillväxt kan den användas som en flexibel utfodringsmodell med en ökad pålitlighet för lokala förhållanden på fiskodlingen. Vid användning i odling kan foderspillet troligen minska och därigenom förbättra både den miljömässiga och ekonomiska hållbarheten.
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