

NORWEGIAN UNIVERSITY OF LIFE SCIENCES





## Summery

In 2011 an experiment was done with juvenile Atlantic Halibut at the on-growing facility of Aga Halibut. Since 2004 the company recorded high mortalities and a low growth rate among the new delivered fish. The cause for that, however, was never specified even after various examinations. Due to that the experimental set up was much aligned, because there no useful indication existed that would have allowed a goal- orientated analysis. Therefore it was decided for a comparison in order to make statements on water quality and possible Bacteria or other microorganism infestation and thus narrowing down the problem.

In the experimental set up at the land-based facility, three different treatments where compared; proteinskimmed water with non - proteinskimmed water and regular feed with antibiotic feed. Overall this was compared to fish which had been directly set out into sea cages.

Regarding the results of the growth and mortality, all three different treatments were insignificant, neither the proteinskimmer nor the antibiotic feed gave any effect on lowering the amount of dead fish as well as any improvement of the growth rate. Only the peak of the mortality curve of the antibiotic feed treatment shifted for about two weeks, and the total mortality amount at the end was slightly lower compared to the other treatments. The results from the sea cage part could not be used, since organization problems prevented any outcome.

However all veterinarian reports plus water-, metal- and chemical analyses made during the years from 2004 until 2011 were collected and in addition all solution attempts were documented and together with the experimental results it was concluded that some form of hidden metal (copper) intoxicification might be the reason behind the high mortalities and low growth rate. Hint to that was given by test of metals in 2007 and the fact that both propeller of the intake water pumps are made out of bronze, containing 80 to 90% copper. In addition up-coming results from a different facility were taken in consideration, confirming the idea of some basic problem within the plant itself.

But due to closing down the land-based facility and moving all the fish into the sea cages, it was not anymore possible to perform a second heavy metal test in order to be positive.

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## Appreviation and Symbols

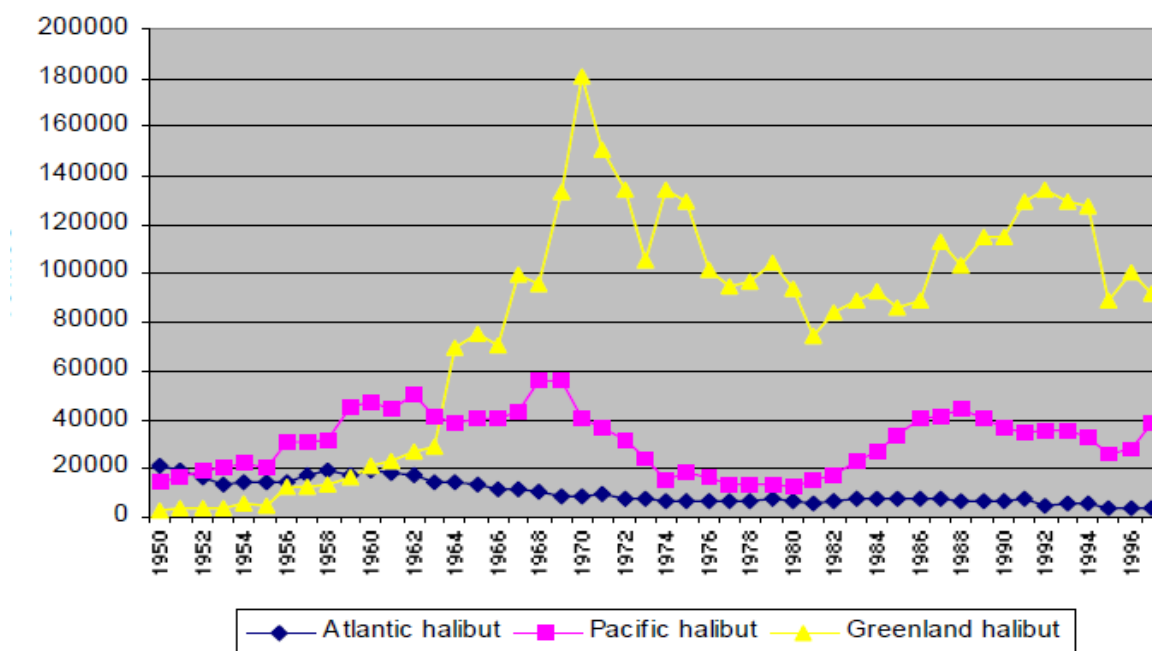
- CAD - Canadian Dollar
- CO<sub>2</sub> - Carbondioxid
- Cu - Copper
- cm - Centimeter
- DHA - Decosahexaenoic acid
- d° - Day degrees
- EHA - Eicosapentaenoic acid
- GBP - Great Britain Pound
- g - Gramm
- i.e. - for example
- h - hour
- Inc. - Incooperation
- IPNV - Infectious pancreatic necrosis virus
- Kg - Kilogram
- Km - Kilometer
- l - Liter
- LPS - Lipopolysaccharide
- m - Meter
- m<sup>2</sup> - Square meter
- m<sup>3</sup> - Cubic meter
- mg - Milligram

mJ - Millijoule  
mm - Millimeter  
Mio - Million  
NH<sub>3</sub> - Ammoniac  
NH<sub>4</sub> - Ammonium  
nm - Nanometer  
NOK - Norwegian Krone  
O<sub>2</sub> - Oxygen  
O<sub>3</sub> -Ozone  
R\* - free Radicals  
Subsp. - Subspecies  
t - Ton  
TGP- Total gas pressure  
UMB -University of Life Sciences  
UK - United Kingdom  
USD - United States Dollar  
UV- Ultra violet  
Zn - Zinc  
°C - Degree Celsius  
> - bigger than  
< - smaller than  
% - Percent  
‰ - Promille



## 1. Introduction

Atlantic Halibut has been steadily declining in the wild since the 1950's - from some 20`000 t/ year down to the current figure of less than 4000 t/ year landings (Figure 1.1). It is now an endangered species due to intense over-fishing, and because they are slow to re-populate, halibut stocks cannot easily recover from the effects of over-fishing [www.list.co.uk]. In 2010, Greenpeace International has added the Atlantic halibut to its seafood red list. The Greenpeace International seafood red list is a list of fish that are commonly sold in supermarkets around the world, and which have a very high risk of being sourced from unsustainable fisheries.



Source: Scotian Halibut Limited, 2009

Figure 1.1 Declining of Atlantic Halibut in the wild since 1950.

Due to its popularity as a food fish, Atlantic halibut has attracted investment in halibut farming. Canada, Norway, the UK, Iceland and Chile are engaged in some form of Atlantic halibut aquaculture production. But the Halibut industry still faces some challenges; inevitable mortalities and the 3:1 ratio of male to female. Females grow 4 to 5 kg in 4-5 years while males consume the same amount of feed but only grow half the size.

Also Aga Halibut, which produces farmed halibut since 1995, is dealing with low growth rate and an above- average mortality of on-growing Halibut. High mortalities around 30 to 40 % are after some time not financially viable, as one dead fish, depending on its size, costs around 40 to 100 NOK, so finding the reason

for the mortalities was of high interest. After several attempts with insufficiently results, the company decided for a scientific investigation.

The aim of the experiment was to figure out if a proteinskimmer can improve the water quality so that it has a positive effect on the growth rate and decline the mortalities for the first and second if the use of antibiotic feed could hint to undiscovered bacteria or any other microorganism at the site.

## 2. Literature Part

### 2.1 Biology of Atlantic halibut

Halibut belongs to the order Flat Fish, Flounder family. Female fish can be over 50 years old, weigh over 300 kg and be 3.5 m long.

Halibut is recorded from the Bay of Biscay Iceland, Svalbard and Novaya Zemlya and from Greenland to the U.S. East Coast (Cape Cod). In Norway, halibut is common along the entire coast.

When they are sexually mature, halibuts gather together on their spawning grounds in 300-700 m deep in ocean streams and fjords. Experiments have shown that halibuts return to the same spawning grounds year after year.

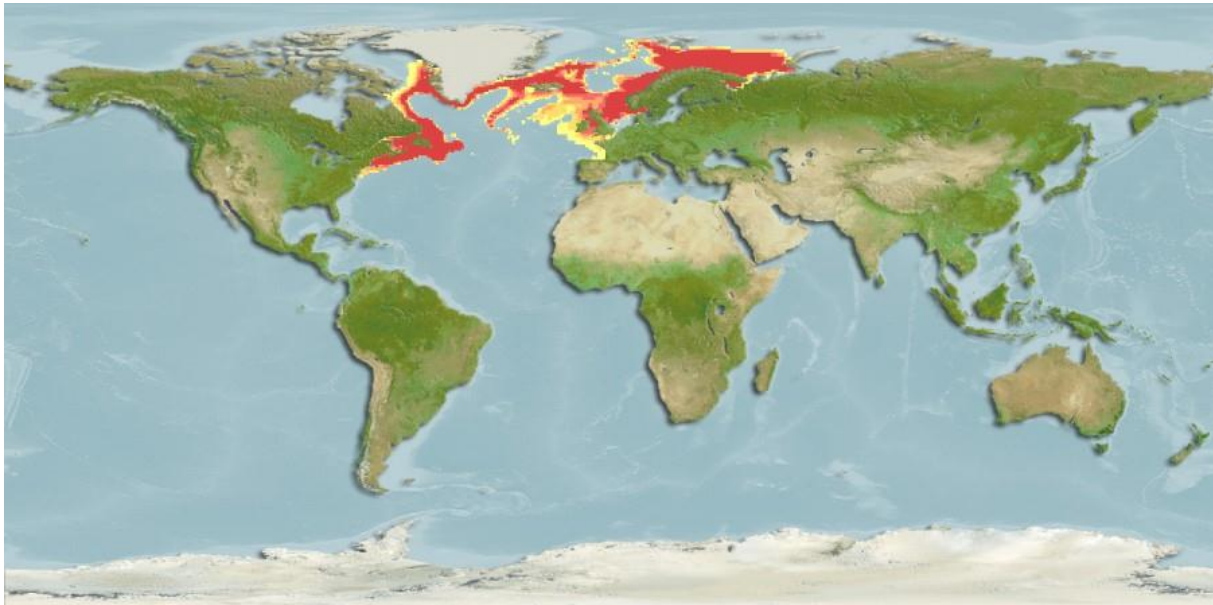
Halibut are batch spawners that produce usually 5-10 batches of 100-200`000 eggs at a 3 to 4 day interval over 1 to 2 months from December to April.

Halibut eggs are about one tenth of the size (volume) of salmon eggs and are adapted for life in deep ocean waters [Foster John, Report 1999 and Fishery and Ocean Canada, 2010]. They can in certain extent regulate their own weight, depending of light exposure. At light exposure the eggs release fresh water and so become heavier and sink. This is one ecological adaptation that is beneficial to prevent the eggs from reaching the surface, because light inhibits hatching, which means that it occurs at night, when the larvae are less visible facing predators and the chance for survival increases.

Halibut larvae are small developed in relation to other fish. The Yolk-sac phase is long, and probably takes place in deep water. First larval food intake occurs in the upper layer of water, with zooplankton [Kveite i oppdrett 2003].

When halibuts are 3 to 4 years old, they begin to migrate out of their nursery areas and can wander long distances (> 100 km).

Large halibut eat almost exclusively fish; especially redfish seem to play an important role. Since redfish are usually on steep slopes and cliffs, the halibut probably out looking to catch them.



Source: [www.fishbase.org](http://www.fishbase.org)

Figure 2.1: Distribution of Atlantic halibut.

## 2.2 History and Development of Atlantic Halibut Farming

The beginning of weaning farmed Atlantic halibut started in Austevoll Norway, 1974. But not until 1988 could a successful production be established. Stolt Sea farm (today: Marine harvest) began a major commercial R&D program in 1988, which encouraged the start of many new Halibut farms in Norway [[www.imr.no](http://www.imr.no)].

## 2.3 Distribution, Production and Market of Halibut Farming

The major players in Europe are Norway, Scotland UK and Iceland.

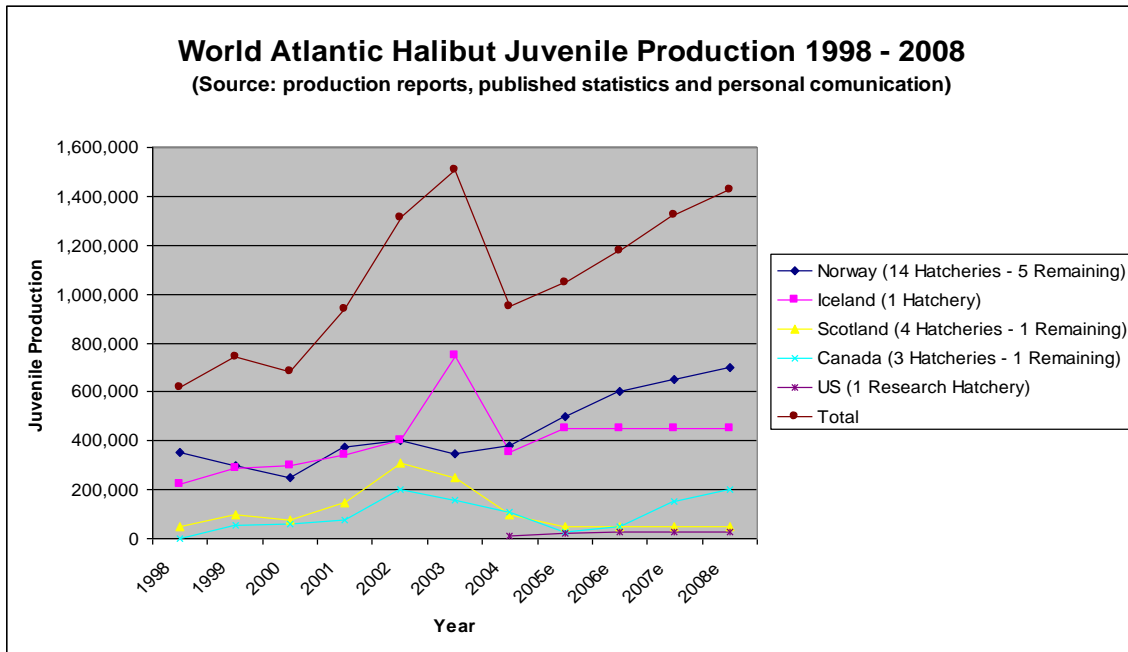
At present UK is left with two companies situated operating in the Shetland Islands with approximately 200 t /year of farmed halibut.

In Norway Marine Harvest is the largest producer followed by Aga Halibut and Nordic Seafarms but also several small operators growing halibut in cages.

In Iceland, Fiskey is the only juvenile producer with 400`000 and 700`000 fish per year. The vast majority is exported to Norway and Scotland, because there is only one small grow-out unit in Iceland.

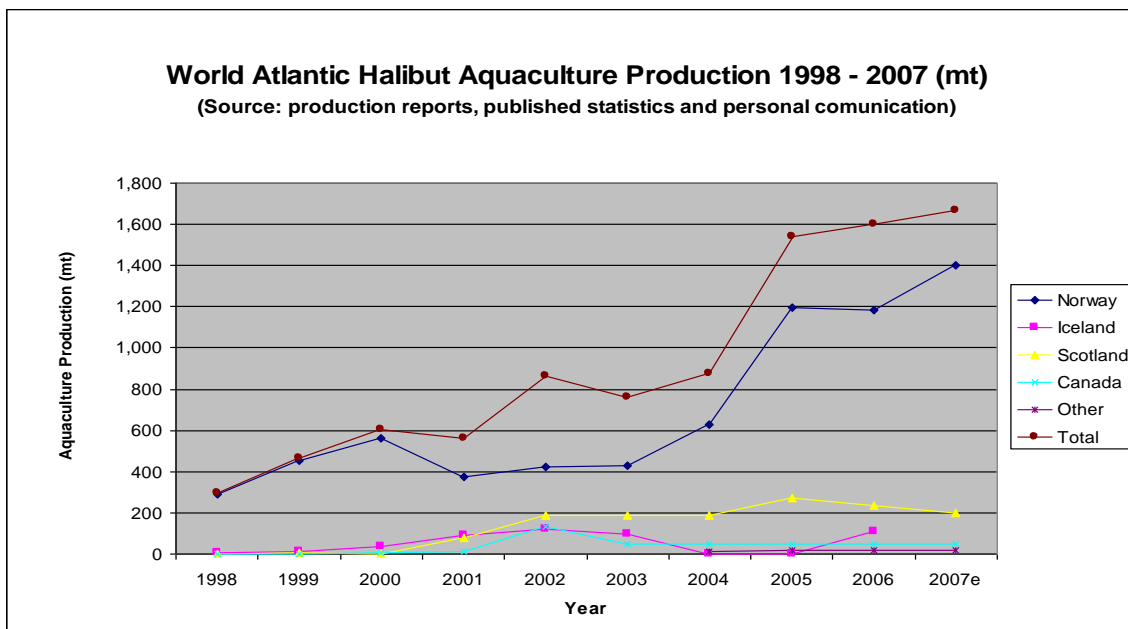
Canadian companies (Halibut PEI, Canaque, Canadian Halibut and Scotian Halibut) are currently (2010/2011) producing around 2600 t/year farmed Atlantic halibut [Annual Statistics, 2009].

Farming of Atlantic Halibut is further practiced in Chile and the west coast of USA. Because research has confirmed that Atlantic halibut females are homogametic and sex-reversed animals, Canada has launched an "All- females" program to improve the growth potential of cultured halibut stocks.



Source: Scotian Halibut Limited, 2009

Figure 2.3.1 Global Production of juvenile Atlantic Halibut (t/year) in the year 1998 until 2008.

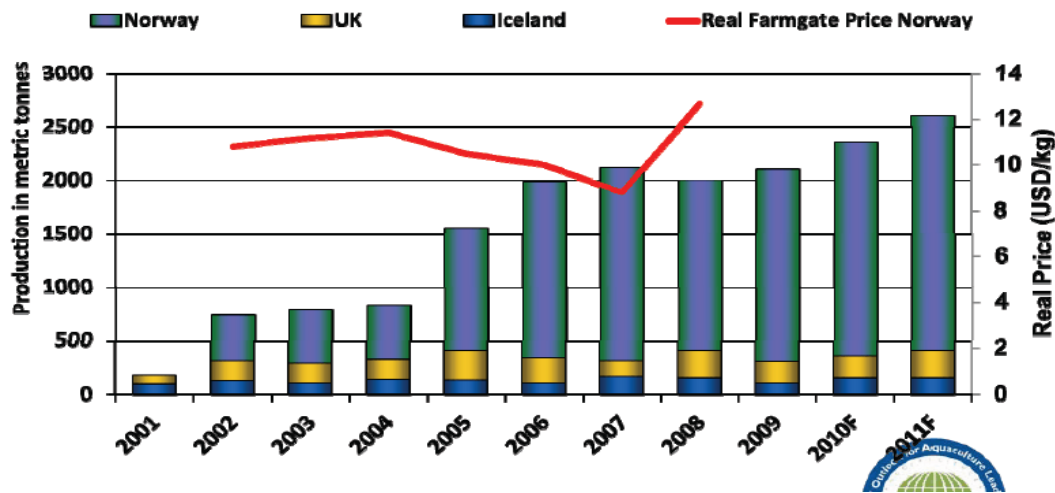


Source: Scotian Halibut Limited, 2009

Figure 2.3.2: Global Production of Atlantic Halibut (t/year) in the year 1998 until 2007.

The total global production (2010) of Atlantic Halibut was around 40`000 t/year, which is far less than the wild catches.

## Atlantic Halibut Production



Source: Kontali-Fish Production Estimates and Trends 2010/2011

Figure 2.3.3: Halibut Farming in Europe.

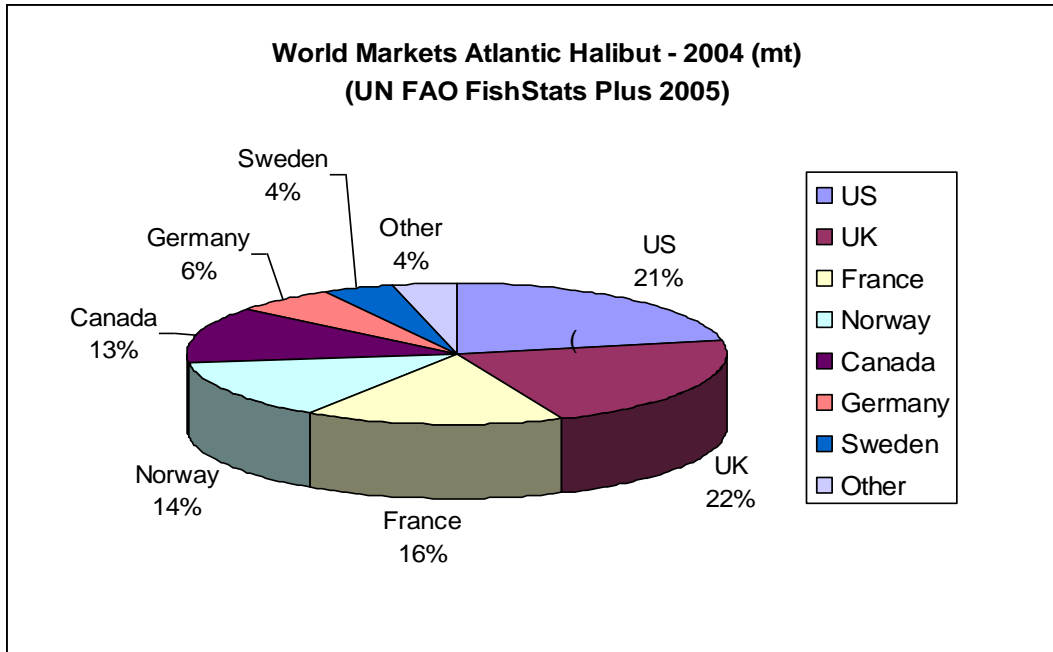
Atlantic Halibut production has steadily increased over the past years but the price maintains high.

### Market

The global and regional “market” for Atlantic halibut is relatively small and presently defined by the availability of supply from the wild fishery.

In 2009 the price of Atlantic halibut achieved a farm-gate price of around GBP 8 /kg (USD 13.04, EUR 9.34) in the United Kingdom and CAD 8.50 (USD 7.45, EUR 5.33) in Canada [seafoodsource.com, Nicky Holmyard, 2009]. Currently (2011) Atlantic Halibut sells for CAD 24 (USD 23.33) per kg [www.fis.com].

Juvenile costs are around GBP 7.50 (USD 12.22, EUR 8.74) to put in the water, and then needs an additional four years worth of feed before it reaches marketable size.



Source: Scotian Halibut 2009

Figure 2.3.4: Market for Atlantic Halibut.

### 2.3.1 Facts and Figures of Norway

At the end of 2007 approx. 70 licensed productions of Atlantic halibut were registered in Norway.

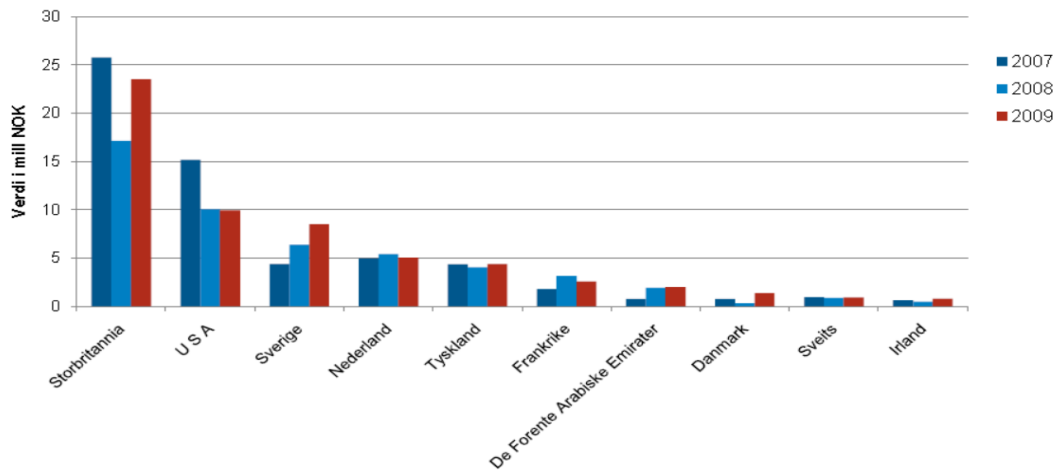
While the average price in 2010 was at 74, 15 NOK/kg is it at present (February 2011) 83, 64 NOK /kg.

In 2010 the export amount was 902 t with a value of 70, 8 Mio NOK which is a 15, 4 % increase to 2009 where 818 t were exported for a value of 61, 3 Mio NOK.

While UK was the largest export market for Norway in recent years (1999 till 2009, see Figure 2.3.1.1), is it to date France with 308 t for 23 Mio NOK (2010) and second UK with 196 t and 13, 7 Mio NOK [www.intrafish.no].

This is most due to the TV and radio campaign “Recette Norvégienne du plaisir”, which basically themes Norwegian seafood in France launched in spring 2010.

### Norsk eksport av oppdrettskveite

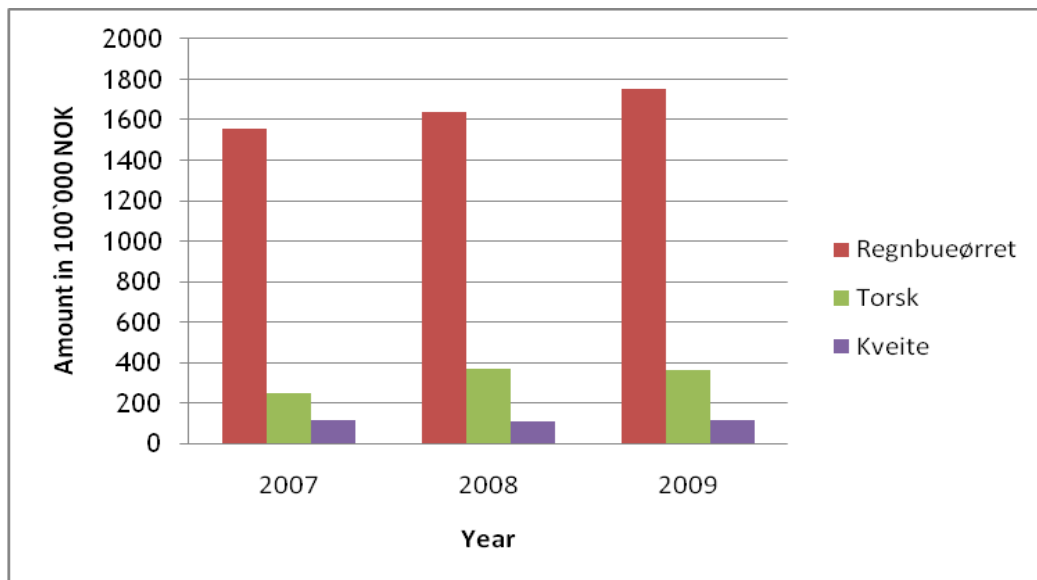


EFF, SSB  
Eksportutvalget for Fisk AS

Eksportutvalget for fisk  
www.seafood.no

Source: www.seafood.no

Figure 2.3.1.1: Norwegian export of farmed Atlantic Halibut from 2007 until 2009.



Source: Statistisk sentralbyrå, www.ssb.no

Figure 2.3.1.2: Sales of slaughtered fish by species in Norway.

Norwegian farmed Atlantic halibut is considered a niche- product, the sales are far below Salmon and other farmed species (see Figure 2.3.1.2), and also because the industry still faces major challenges; the inevitable mortalities and the 3:1 ratio of male to female juveniles. Females grow to a marketable size of 4 to 5 kg in 4 to 5 years, while male fish consume the same amount of feed but only grow to half the size.

## **Norwegian Producers**

### ***Nordic Seafarms***

Nordic Seafarm AS was established in 1995 and today consists of Norsk Kveite AS, Halibut AS, Norsk Kveiteavl AS and Nordic Halibut AS, which is also their trade mark.

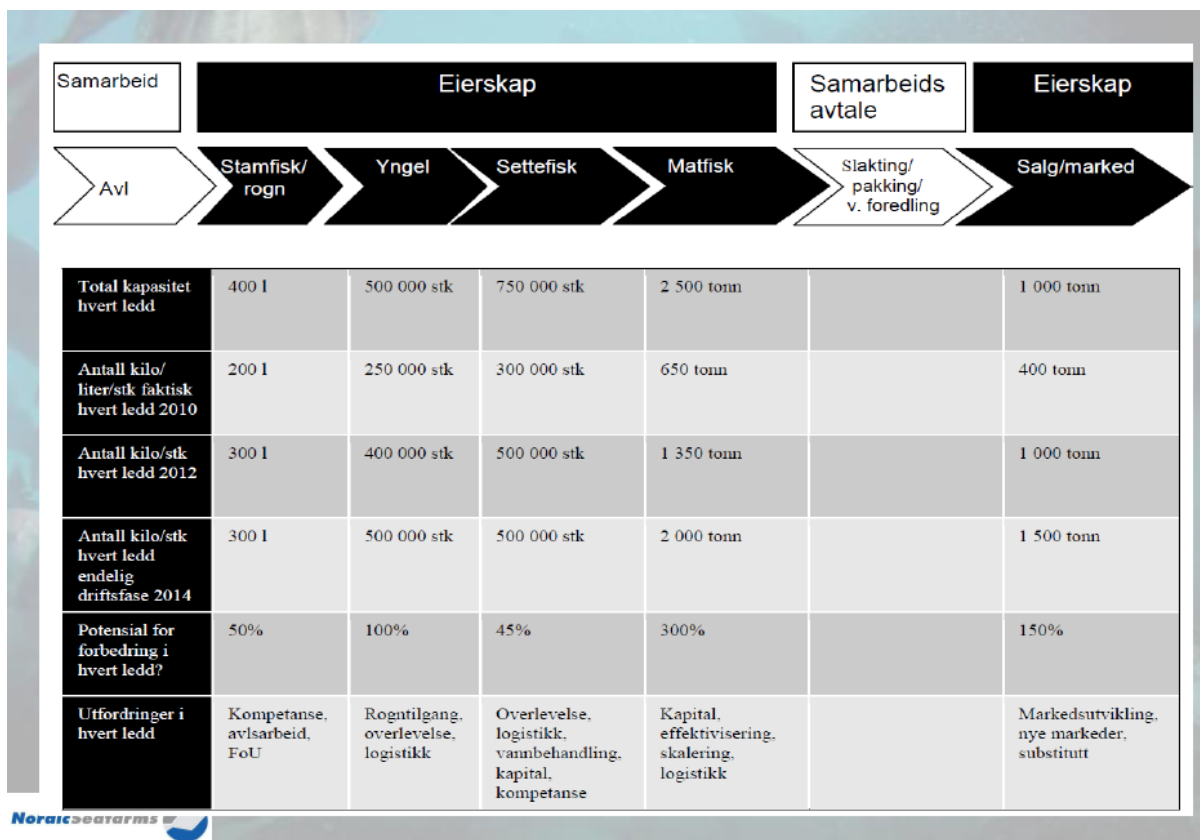
The total stock of Halibut in the Nordic Seafarms was 187,000 individuals of different age classes at the end of August 2010, with a total biomass of about 560 t. More than half of the biomass is in sea cages; the rest is in the plant at Averøy [Nordic Seafarms ASA].

Nordic Seafarms has 356 broodstock fish located in Midsund i Møre and Romsdal, and in addition to their own hatchery, are working closely with Brandal Aquaculture AS, Norway's largest producer of halibut juveniles. Approximately 50`000 juveniles come from it and ca 20`000 are self-produced fry plus some minor amounts from other producers.

The total harvest volume for 2010 was between 400 and 450 t with prices around 85-90 NOK per kg and an average slaughter weight of 6 kg. The main market is UK, with Norway as number two[[www.nordicseafarms.no](http://www.nordicseafarms.no)].

The company owns 50 % of Maritime Marine Aquaculture Inc. (MMI) resident in New Brunswick. MMI has been a positive development in the production of fry and income for the last couple of years.





Source: nordicseafarms.com

Figure 2.3.1.3: Present status and planning of production of Atlantic Halibut by Nordic Seafarm.

### Marina Harvest

Sterling White Halibut is a branch industry of Marine Harvest cold water species. The fish is farmed on several locations, in the middle of Norway in Nord Trøndelag County, where the larvae are produced. On-growing fish are in Rogaland County near Stavanger.

Marine Harvest Norway harvested totally 60,000 metric tons, followed by Scotland at 10`000 metric tons, Canada at 9`000 metric tons and Chile at 7`000 metric tons in 2009 [seafoodsource.com ,January 2011]. In 2009 Sterling White Halibut had a turnover of 62 Mio NOK.

## 2.4 Production stages and Farm practises

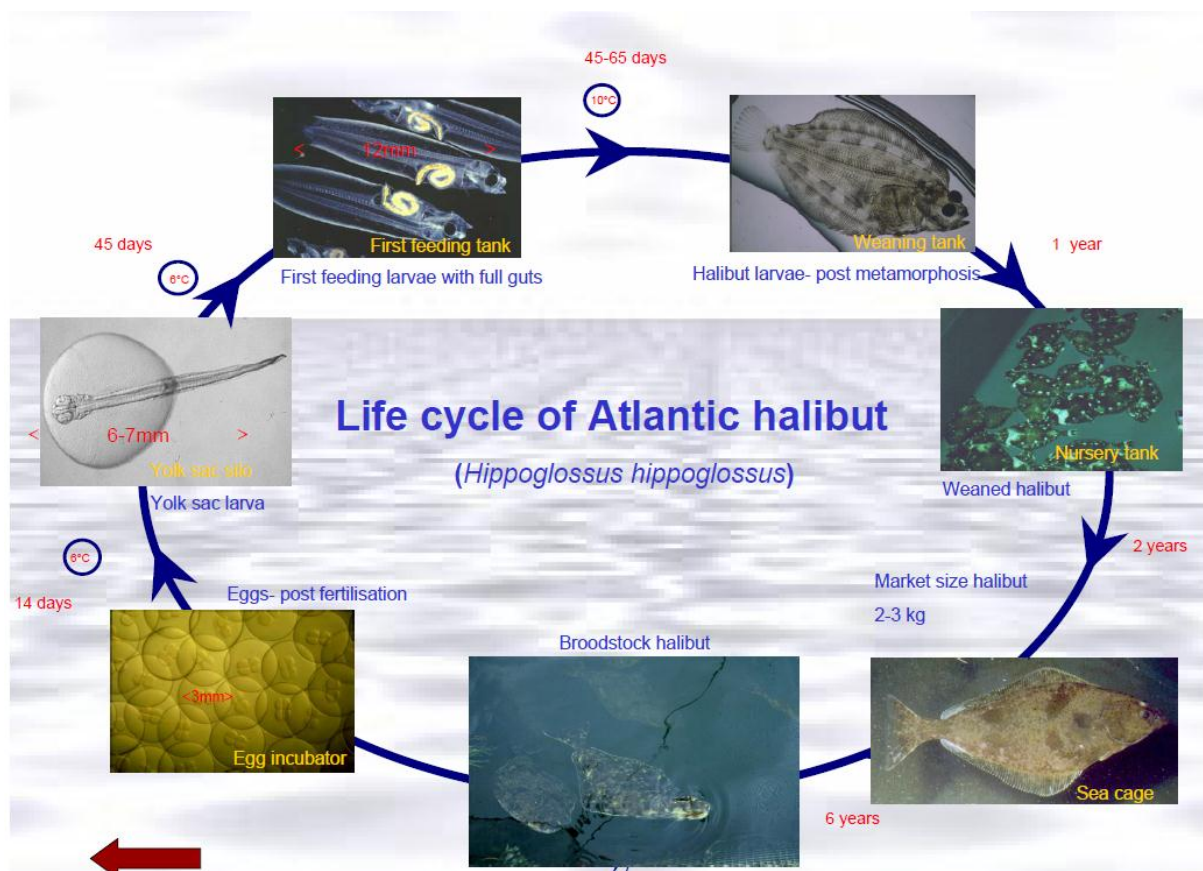
The key stages in halibut aquaculture are (see also Figure 2.4.1):

### Hatchery

- Broodstock and spawning
- Egg incubation
- Yolk sac larvae development
- First feeding
- Metamorphosis
- Weaning

### Nursery and On-growing

- Landbased facility
- Sea cages



Source: Seafish industry  
Figure 2.4.1: Production stages of Atlantic Halibut.

## 2.4.1 Broodstock and Hatchery

### Broodstock and Incubation

Until recently brood-stock fish used to be caught wild halibut but lots of disadvantages like reduced appetite, parasites and damages on the fish led to the establishment of farmed brood-stock and breeding station, which not only reduces the risk of diseases and infections but also enables selection of the best fish and the delivery of certain egg amount throughout the year.

The natural spawning season for Atlantic halibut is from February - May, but water temperature and light can vary under culture conditions, so that some fish are ready to produce eggs at all times of the year [Forster John, report 1999]. Age at sexual maturity in farming is about 2 years for males and 4 -5 years for females. Eggs and milt are stripped out manually and eggs are fertilized immediately after stripping and incubated in up-welling incubator (Figure 2.4.1.1) in a dark room, because light adversely affects development and hatching. Since they are adapted for life at depths of 1000 and below, it is not surprising that the eggs are negatively affected by light and need constant cool incubation temperature of 4-6 °C as well as full strength salinity water. Incubation systems vary in Norway, 250 l containers are generally used with water flows between 1-2 l / minute.

About 10 days (65-75 d°) after fertilization, the eggs are disinfected and transferred to the silos (Figure 2.4.1.1) where hatching and the yolk sac phase takes place.

Normally, it is expected that at least 90 % of the eggs will be fertilized and that 75 - 80 % of the fertilized eggs will hatch [Forster John 1999].



Source: Seafish industry

Figure 2.4.1.1: Conical -shaped up-welling incubation vessels (450 l, left picture) and larval rearing tanks small (1000 l, right picture), equipped with flow-, temperature control.

### *Yolk sac larvae development*

Compared to other marine fish, the yolk sac stage in halibut is very long. The larvae feed on the yolk sac over 40 days. Water temperature is between 7-9°C and the fish are kept in darkness.

Halibut larvae are adapted to very stable physical conditions with no abrasion and low levels of micro-organisms, therefore the larvae are hold in silo shaped tanks up to 15`000 l with an inflow of new water at the bottom and an outflow at the top. Also much smaller tanks are used, ranging in volume from 500 - 2`000 l, with this facilitates temperature control is better suited for smaller egg batches. Survival during the yolk sac stage generally ranges between 50-70 % and the larvae grow from 6 to 12 mm long [Wray 1998]



Source: Austevoll Havbruksstasjon - [www.fiskeoppdrett.no](http://www.fiskeoppdrett.no)

Figure 2.4.1.2: Halibut larvae - from yolk sac to start-feeding phase.

### *Startfeeding*

The larvae are first fed 30 to 50 days (260-270 d°) after fertilization and the larvae are transferred to shallow, circular tanks. They are removed from their silos by drawing them to the top with light and scooping them out with buckets.

Water temperature gently rises from 6°C to 8°C and after transferring increase slowly up to 10 - 12°C [kveitemanualen.imr.no].

Feed is usually enriched live Artemia or filtered copepods from the sea. Artemia can only be used successfully as a first-feed for halibut larvae if its nutritional

composition is supplemented with enrichment of additional nutrients, in particular with essential amino- and polyunsaturated fatty acids. Enrichment aims to simulate the nutrition provided by copepods or by the natural zooplankton diet of wild halibut. Enrichment procedures for Artemia show considerable variation among hatcheries because the detailed nutrient requirements of first-feeding halibut are not fully known. In a number of hatcheries the Artemia are enriched with mixtures of Super Selco (INVE Aquaculture, Belgium) and a marine heterotroph Algamac 2000 (Aquafauna Biomarine Inc., USA). These are then added to the tanks at the rate of 1000 Artemia nauplii/L twice a day; this corresponds to a daily ingestion rate for each halibut larva of approximately 2000-3000 prey organisms/day [seafish industry.com].

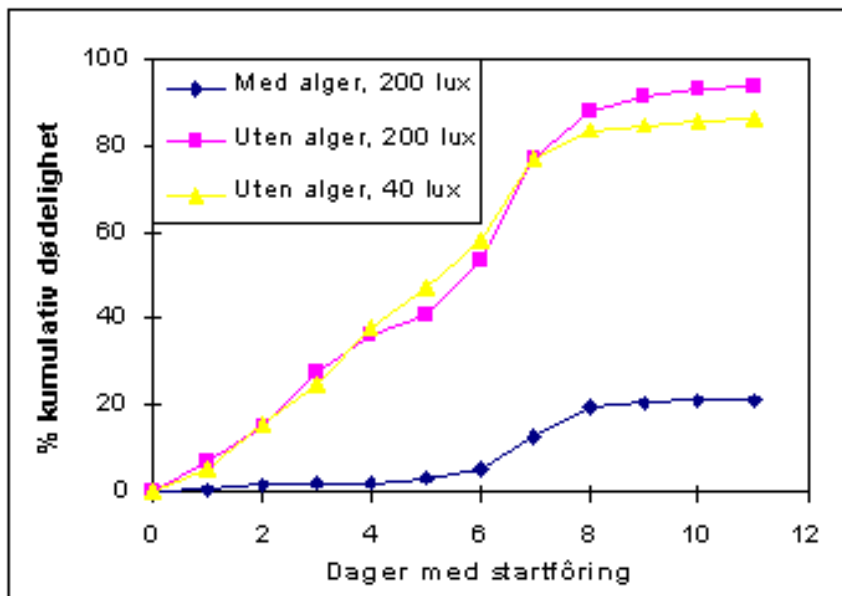
In hatcheries where the Artemia is supplemented with 20% copepods, 30% more survival through start-feeding and 90% fully pigmented fish are achieved. Compared to Artemia and rotifers, copepods have higher amount of DHA as well as a higher ratio of phospholipids than triglyceride. Triglycerides are better digestible and also necessary for marine larvae since their having a low *de novo* synthesis [Webster and Lim 2002].

“Green water” / Algae water

Before the larvae are stocked, the start-feeding tanks are “greened” with appropriate microalgae at concentrations of up to 10<sup>7</sup> algal cells/l; these concentrations are maintained by further daily additions of algae up to approximately 500 d° post-hatching. Figure 2.4.1.3 shows the accumulated mortality of the larvae after days of start-feeding. Algae water has a dramatic enhancing effect on survival among larvae in the first days after transfer to first feeding tanks. The mortality rises after day 6 with larvae that have not fed. The lowest mortality occurs when algae were in the tanks.

There has been some debate within the aquaculture community over the mechanism by which green water leads to increased feeding, which is merely due to the physical presence of the algae. Inert particles can successfully substitute for micro-algae. Further experiments indicated that improved feeding is likely to result from slight turbidity improving contrast between the background and the *Artemia* sp. nauplii on which halibut first feed [Leahey 2008].





Source: [www.kveitemanualen.imr.no](http://www.kveitemanualen.imr.no)

Figure 2.4.1.3: Mortality with different environmental conditions.

### Metamorphosis

During metamorphosis the left eye wanders over to the right site. The larvae are flat, pigmented on the eye side and lay on the non pigmented side down at the bottom. This extraordinary biological change usually occurs around 90 days after hatching, depending on the rate of larval growth.

False Pigmentation (Figure 2.4.1.4) and incomplete metamorphosis are two common aesthetic differences in relation to wild fish. The white color is gradually replaced by a gray color, while the dark blind side remains dark. The importance of these aesthetic differences for future growth and survival is not clear; it could indicate that it has little or no effect on growth. The proportion of fish that is reversed compared to normal have so far no effect on growth or survival [Kveite i optrett 2003].

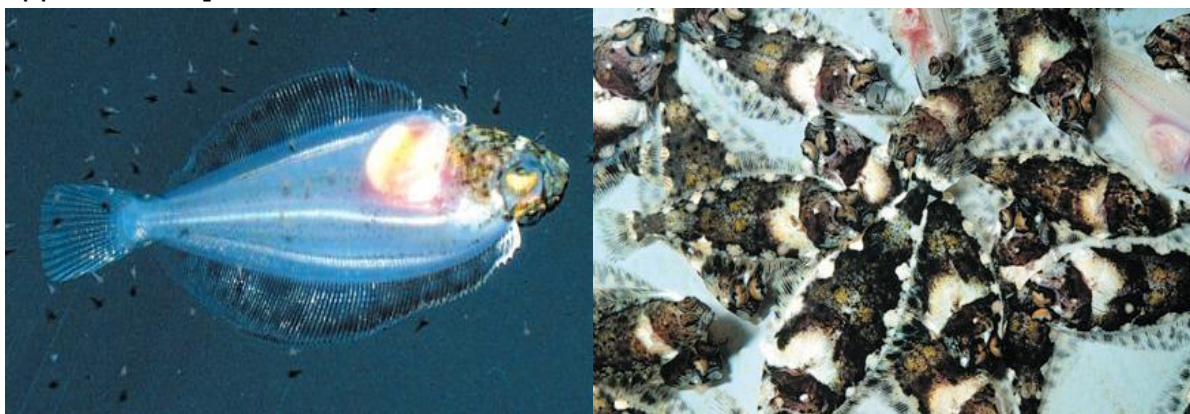


Foto: Austevoll Havbruksstasjon -[www.fiskeoppdrett.no](http://www.fiskeoppdrett.no)

Figure 2.4.1.4: False pigmented and normal pigmented halibut fry.

### *Weaning*

Weaning occurs when the diet of the newly metamorphosed juveniles is changed from live feed to formula feeds. Weaning tanks (Figure 2.4.1.5) are small in order to have control over the fish and maintenance can be done manually. During this process the young fish, weighing between 250 - 300 mg, are offered both types of feed, with the supply of live feed being gradually reduced. For weaning, the water temperature is increased to 13-14°C, which has been shown to be optimum for growth of young halibut. This process is usually complete in 30 days; by which time the fish weigh about 1 g. Expected survival is about 70 % [Pittman, unpublished].



Source: Seafish industry

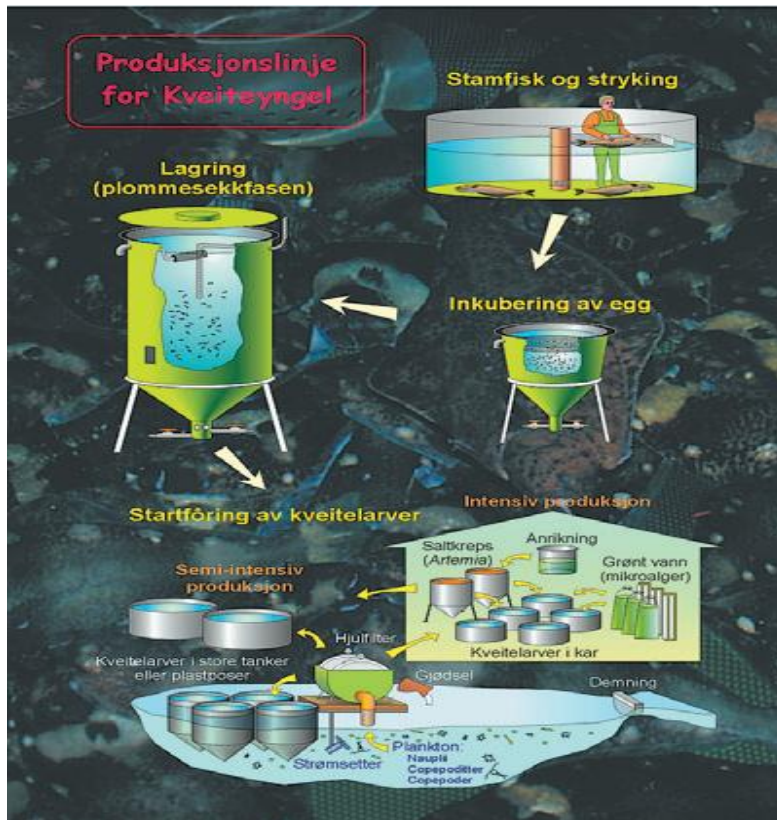
Figure 2.4.1.5: Halibut weaning and nursery tanks equipped with temperature control and aeration.

### *Nursery*

If optimum temperatures are maintained, halibut will reach a weight of 5-10 g in 150 days after hatching and 150-200 g by the end of their first year. This stage's length of time varies a lot, because the purpose is to hold and grow the young halibut until they can be moved or sold, to an on-growing system.

For example, if the young halibut are to be on-grown in cages, it is usually thought necessary to grow them in the nursery to 250 - 500 g. If they are to be on-grown in land based tanks, transfer can be made much earlier at, around 10-20 g [Forster 1999, Seafish industry 2003 and Kveite i oppdrett 2003].

In Figure 2.4.1.6 production stages from brood-stock until start-feeding are all summarized.



Source: Terje van der Meeren, Havforskningsinstituttet- [www.fiskeoppdrett.no](http://www.fiskeoppdrett.no)

Figure 2.4.1.6: Production stages for Halibut-fry. Halibut development through the early stages is divided into: egg stage, yolk sac phase and start-feeding of larvae.

## 2.4.2 On-growing

When the fry are big enough to be purchased and exposed to either

- 1) Land based facilities
- 2) Sea cages

### 1) Land-based facilities

Halibut are naturally docile and not easily to be stressed, therefore they tolerate crowding well and stocking densities are usually expressed in terms of kilograms per square meter (Figure 2.4.2.1). If the fish in tanks on land set there are no requirements for minimum size, but it is customary to set out the fish between 5 and 10 g. If the fish are put into the sea, it is recommended to do this in spring, with a size of 200 g.

Halibut tanks must be flat so that the fish can rest between feeding times. Both the density and size in the tanks affect growth. The installation of shelves in the tank increased area and density per shelf is reduced.



Halibut likes to dig into, and often have damages in tanks without substrate. It is therefore recommended to use substrate such as gravel or plastic mesh. Halibut dislike bright light but a little light is important for several reasons. It has been concluded that there should be about 18 hours light per day, and the rest of the day with dimmed light [kveitemanualen.imr.no].

Table 2.4.2.1: Recommended maximum stocking densities for halibut (Englesen 1995).

| Fish weight (g) | Density (kg /m <sup>2</sup> ) |
|-----------------|-------------------------------|
| 2 - 149         | 10                            |
| 149 - 448       | 20                            |
| 448 - 1495      | 30                            |
| 1495 - 2496     | 40                            |
| 2496 - 4264     | 50                            |
| 4264 - 6410     | 60                            |
| 6410 +          | 70                            |

Source: Report Forster 1999

### **Temperature**

The optimum temperature for growth in halibut should be in the range of > 4°C and < 14°C, according to Englesen( 1995).

Table 2.4.2.2: Optimum temperature ranges for the growth of halibut at different sizes (Englesen, 1995).

| Fish weight (g) | Temperature °C |
|-----------------|----------------|
| 2-25            | 11-14          |
| 25-100          | 11-13          |
| 100-500         | 10-12          |
| 500-1000        | 9-11           |
| > 1000          | 7-11           |

Source: Report Forster 1999

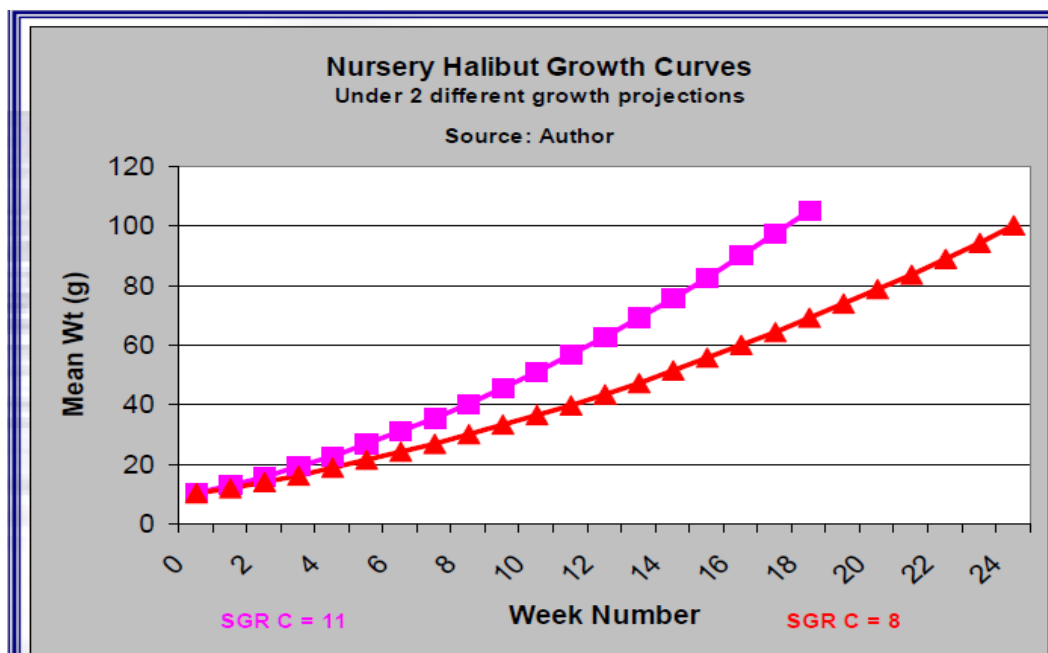
To ensure near maximal growth and near optimal growth efficiency and high survival the following temperatures are recommended:

12-13°C for 10-60 g juveniles,

8-11°C for 100-500 g juveniles and

6-9°C for 3-5 kg immature halibut.

But with increasing temperature there is also a tendency for increased mortality within in the fish [Bjørnson 1995].



Source: Seafish industry

Figure 2.4.2.1: Growth rates at different temperatures (11°C and 8°C).

Growth rate is influenced by many parameters but temperature seems to be one of the most deciding factors (Figure 2.4.2.1).

According to several studies juvenile halibut exposed to higher temperatures (within the optimum) showed a periodical higher growth rate than fish held in lower temperatures during the same period, but overall both groups (higher and lower temperature) showed the same growth rate in the end of the observation. Halibut apparently show the same compensatory growth as other farmed animals. Halibut appears to be eurythermal, which is expressed by high growth rates over a relatively wide temperature rang [Jonassen et al. 1999]. This characteristic is also size related indicating that thermal optimum of halibut decrease as the fish increase in size, so that the growth of big fish is not as much influenced by temperature then for small fish. In other words temperature tolerance increases with size [Jonassen et al. 1999].

Changes in temperature from 7.5 °C up or down by 3 °C gave only a short-term effect on appetite. This indicates that rapid changes in temperature do not have long-lasting effect [www.imr.no, Institute of marine research].

## Oxygen

Halibuts tolerance limits for oxygen is assumed to be much equivalent to other marine species. When oxygen saturation drops approximately < 80 % the fish are likely to reduce growth and mortality will occur when the saturation is approximately < 30 %.

Mild over saturation of oxygen is probably not harmful, since addition of oxygen in the vessel has not created problems [www.imr.no]. Higher temperatures increase the oxygen consumption (Table 2.4.3.3).

Table 2.4.3.3: Oxygen consumption (mg O<sub>2</sub>/ kg / min) for different sizes of Atlantic halibut and different temperatures.

| Size     | Temperature (C) | Oxygen consumption (mg/kg/min) | Source                    |
|----------|-----------------|--------------------------------|---------------------------|
| 10-80 g  | 6               | 1,9                            | Jonassen m.fl. (2000)     |
| 20-100 g | 7               | 2,3                            | Hallaråker m.fl. (1995)   |
|          | 10              | 2,7                            | 7 Hallaråker m.fl. (1995) |
|          | 12              | 2,3                            | Jonassen m.fl. (2000)     |
|          | 13              | 3,0                            | Hallaråker m.fl. (1995)   |
|          | 16              | 3,3                            | Hallaråker m.fl. (1995)   |
| 1-4 kg   | 8               | 1,4                            | Rønnestad (1988)          |

Source: [www.imr.no](http://www.imr.no)- Institute of marine research

Table below (2.4.3.4) gives an orientation over the Oxygen saturation in sea water. Marine species are exposed to lower oxygen saturation then fresh water species and therefore tolerate less oxygen saturation.

Table 2.4.3.4: Oxygen per liter of sea water with 100 % saturation and different temperatures (Salinity 35 ‰).

| Temperatur ( °C) | Oxygen (mg/l) |
|------------------|---------------|
| 4                | 10,5          |
| 6                | 9,8           |
| 8                | 9,5           |
| 10               | 9,0           |
| 12               | 8,6           |
| 14               | 8,2           |
| 16               | 8,0           |

Source: [www.imr.no](http://www.imr.no)- Institute of marine research

## 2) Sea cages

Halibut cage farming can be an alternative to land-based aquaculture. Cage

farming provides a much lower investment than land-based aquaculture and is therefore easier to establish, but preliminary estimates indicate that the production rate can reach the same level [Kveite i merd 97/98].

Today`s halibut sea farming is based on technology from salmon farming (cages, feeding systems) that adapted for halibut needs and provides little control of the fish compared to onshore facilities, and additional monitoring is necessary like underwater camera. It is likely that unexplained loss of individuals will be greater in a cage system than a land-based facilities [Kveite i merd 97/98].

The cage location should be placed on sites where the surface temperature varies as little as possible. Temperature fluctuations in the sea are much higher near the surface than further down .The surface layers are often higher (summer) or lower (winter) temperatures than water only a few meters further down. A major kill of farmed halibut occurred in 1997 when water temperatures at a net pen farm at Austevoll, Norway exceeded 18 °C, lending further support to the idea that on-shore systems are the best way to farm on- growing halibut [Forster 1999].

Cages can ranges from 5 m to 30 m depth and require a flat bottom that fish can lay on to. As biomass increases shelves are usually used to provide more space and allow higher stocking within one cage. Stocking density usually reaches from 20 kg/m<sup>2</sup> to maximal 50 kg/m<sup>2</sup>.

Feed cameras are used to observe feeding habits and to control feed amount.

Regarding the mesh size of the nets it depends very much on the fish size; too small will create fouling and too big can be a problem with predators and other fish intruding.

Dead fish has to be removed weekly using either divers or ROV (remote operated vehicle)-technology to separate dead from laying fish alive on the bottom, which can be very costly but supporting technology is missing [www.seafish.com, 2002].

The production of harvestable fish is long and may take 3-4 years from 300 g to 6 kg. By using large fry (e.g. between 300 - 600 g) difficult sorting could be reduced and also reduce production time in the plant.

### **Salinity**

Halibut is stenohalin (tolerate little variation in salinity), and has little fluctuation in salinity within the range [www.imr.no]. Salinity in the coastal current is usually between 33 ‰ and 35 ‰, but may be lower in fjords with high freshwater runoff.

Recent results indicate that salinity down by 15 ‰ does not have any negative effects [Opstad and Rust 2004]. In experiments with juvenile Atlantic Halibut run by Imsland et al. 2008, it was found that a lower salinity of 15 ‰ resulted in a better SGR of 1, 28 % and feed conversion efficiency (FCE) of 1, 21 compared to a salinity of 32 ‰ with SGR of 1,16 % and FCE of 0,97.

### 2.4.3 Feed, Feeding frequency and Growth

Feed composition is important regarding the conversion into muscle mass of the fish. The better adapted the feed is to the fish`s needs, the larger the proportion recovered as growth. Knowledge of macro-nutrients (i.e. protein, fat and carbohydrates) is rather incomplete.

Halibut have a high requirement for protein in accordance with other marine carnivorous fish [Anders et al. 1996].

#### *Proteins*

The optimal dietary protein level for Atlantic halibut depends on body size [Grisdale-Helland & Helland, 2002]. Experiments show that small halibut (7 to 180 g ) respond with increased weight gain when dietary protein increases from 41 % to 62 % and carbohydrate level decreases from 27 % to 3 %. However Halibut grown from 150g to 550g did not result in increased growth when dietary protein was over 51 %. And when halibut reached 600g and bigger the growth was not improved by protein levels greater than 37 %.

The digestibility of protein in dry feeds is in the range of 82 % to 86 %. Use of the higher carbohydrate level gives greater fecal organic matter loss to the environment and decreases the total digestibility [Grisdale-Helland & Helland 1998].

#### *Fats*

DHA 22:6n-3 and EPA 20:5n-3 are essential fatty acids in diets for marine fish. The lack of  $\Delta$ -5 desaturase activity in marine fish prevent the conversion of linoleic acid (18:2n-6) to AA (20:4n-6), which is essential for producing EPA [Webster and Lim 2002].

Higher dietary lipid levels (around 30 %) increase the storage of fat in the tissue and halibut has its fat depots close to the bones and fins and reduces growth [Anders et al. 1996].

#### *Feeding frequencies and Growth*

Limited information are available concerning optimal feeding frequency or the optimal feeding times necessary to maximize growth in juvenile halibut [Berge and Storebakken 1991]. Juvenile halibut (~ 20 g) fed more than three times per day had a SGR from 1.6% to 2.0%. According to Schnaittacher et al. 2005; 20 g halibut being fed 5 times a day show significant greater final weights instead of feeding frequency of 1 times day.

In contrast, the size variation of larger halibut (140 g) fed once per day under a variety of temperature regimens (5.0-14.9 °C) greatly increased over the course of the experiment and the SGR decreased to 0.55 % [Bjørnsson & Tryggvadottir 1996]; however, SGR values always decrease as fish increase in size [Bjørnsson 1995].

Table 2.4.3 gives an overview over the specific growth rates at different temperatures and different sizes. Smaller fish show higher growth rate than bigger fish and also higher temperatures improve feed uptake and thereby the growth rate. But the bigger the fish get, the less are they responding to temperature variation [Grisdale-Helland, 1998], which indicates that they have a wider temperature tolerance and will show the same appétit in e.g. 12 °C and 10 °C, whereas with smaller fish appétit will reduce when changing the temperatures for about 2 degrees.

Table 2.4.3 Specific growth rate (SGR %/day) reported from different growth experiments with halibut, with different fish sizes and at different temperatures.

| Size(g) | Temperature |          |         |         |         |     |
|---------|-------------|----------|---------|---------|---------|-----|
|         | 6           | 8        | 10      | 12      | 14      | 16  |
| 10      | 1,0-1,3     | 1,6-2,0  | 1,8-2,5 | 2,4-2,9 | 3,1     | 3,0 |
| 50      | 0,6-0,7     | 0,8-1,25 | 1,0-1,5 | 1,2-1,8 | 1,4-1,7 | 1,6 |
| 100     | 0,5         | 0,6      | 0,8     | 0,8-1,7 | 1,5-2,1 |     |
| 200     | 0,3         | 0,4-0,7  | 0,8-1,0 | 0,7-0,9 | 0,8     |     |
| 500     | 0,2         | 0,2-0,5  | 0,7     | 0,8     |         |     |
| 1000    | 0,2         | 0,2      | 0,3     | 0,4     |         |     |
| 2500    | 0,1         | 0,4      | 0,6     | 0,3     |         |     |
| 5000    | 0,2         |          |         |         |         |     |

Source: www.imr.no- Institute of marine research

With temperatures below 5°C growth stagnates more or less.

#### 2.4.4 Water treatment

##### UV

UV light is comprised of electromagnetic radiation of wavelengths ranging from 10 nm to 400 nanometers (nm).

UV-A (Long Wave UV): 315-400 nm

UV-B (Middle Wave UV): 280-315 nm

UV-C (Short Wave UV): 200-280 nm

Vacuum UV: 10-200 nm

## UV for disinfection

UV disinfects at a wavelength of 254 nm by penetrating the cell wall of the microorganism. The amount of UV delivered to the organism is called the intensity. The UV energy permanently alters the DNA structure of the microorganism in a process called thymine dimerisation.

This inactivates the microorganism and renders it unable to reproduce or infect.

Advantages of using UV it inactivates microorganism within seconds and it does not alter water chemistry and its constituents such as pH, taste, odor, color etc.

| UV Doses for Inactivation of Selected Fish Disease Causal Pathogens |                               |
|---|-------------------------------|
| Pathogen  | UV Dose (mJ/cm <sup>2</sup> ) |
| <b>Bacteria</b>   |                               |
| <i>Aeromonas hydrophila</i>   | 22,1                          |
| <i>A. Salmonicida</i>   | 13,1 to 29,4                  |
| <i>Pseudomonas flourescens</i>                                      | 13,1 to 29,4                  |
| <b>Viruses</b>  |                               |
| VHS   | 5,0*                          |
| ISA   | 120,0                         |
| IHNV (RTTO)   | 30,0                          |
| IPNV (Buhl)   | 150,0                         |
| <b>Protozoa</b>   |                               |
| <i>Ichthyophthirius tomites</i>                                     | >300,0                        |
| <i>Myxosoma cerebralis</i> (Whirling Disease)                       | 40,0**                        |
| <b>Fungii</b>   |                               |
| <i>Saprolegnia zoospores</i>  | 39,6                          |

Source: www.thefishsite.com

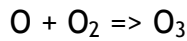
Figure 2.4.4.1: Doses needed for specific fish diseases found in aquaculture systems.

UV is affected by a number of different factors including UV transmittance, turbidity, hardness and suspended solids, it will all reduce intensity as well as a fast flow rate, will lower the UV dose, whilst a slow rate will increase it.

## Ozone

Ozone is formed by an O<sub>2</sub> - molecule dissociates to free radicals and then reacts with another O<sub>2</sub> - molecule to O<sub>3</sub>.





### *Effect in Saltwater*

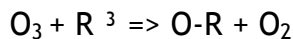
In seawater  $O_3$  decomposes within minutes or seconds and it will form reactive radicals, including OH. The amount of radicals increases with increasing ozone concentration in the water and increasing pH. It is not only the redox-potential which is essential for the reactions that take place, but also the concentration of reaction partners and the rate constants for reactions.

Significant is that hydrobromit react with ozone back to bromide. Practical experience has shown that one should carefully ozone seawater, i.e. not around 350 mV to avoid the formation of the stable bromate.

Bromate is reactive, but is also described to have highly toxic effects on aquatic organisms. It also has the property that it builds up in the recycled plant using ozone.

The driving force for the transfer of ozone from gas phase to water phase is proportional to the shared pressure of ozone in the gas phase.

Ozone seems strongly oxidizing i.e. one of the  $O_2$  atoms easily split off to Ozone and bind to other components in the water.



Ozone is often used as a disinfectant in aquaculture facilities, however residual ozone must be removed before it comes into contact with fish.

A short UV wavelength of 254 nm consumes residual ozone. Ozone absorbs the UV energy and quickly dissipates, breaking down in  $O_2$  molecules.

Typically, 1.0 g/l of ozone can be removed with a UV dosage of 90  $mJ/cm^2$  [www.thefishsite.com, August 2010].

In Experiments it was shown that UV-light efficiently inactivated bacteria, while the IPN-virus was not very susceptible. However, bacteria associated to particles survived high doses of UV-light. Good results were achieved when the water was filtered through a cover with a pore size of 50 or 80  $\mu m$  before the UV-treatment. Low concentrations of ozone (0, 15-0, 20 mg/l) were effective for the inactivation of both bacteria and virus after 1 minute in fresh water, brackish- and seawater. Ozonation is generally a more expensive and complicated disinfection process than UV-irradiation.



However bacteria, which had been treated with UV-light, showed a surprising ability to repair their UV- damage under subsequent normal conditions. A phenomena studied was the so-called photo-reactivation, which is the ability of bacteria to repair UV-damage by the help of enzymes at visible light, e.g. light of a lamp or sunlight. Due to this effect, it is necessary to increase the UV-dose by 3-4 times compared to the inactivation without repair [www.aquaflow.org].

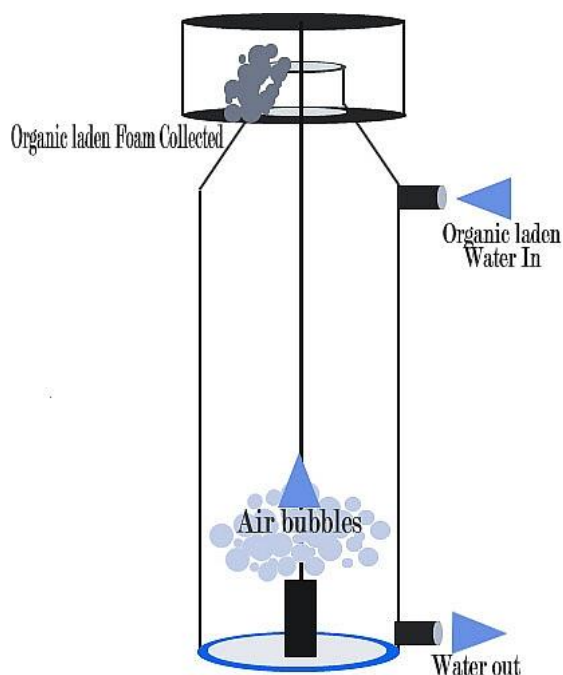
### Proteinskimmer

A protein skimmer or foam fractionators is a device used mostly in saltwater to remove organic compounds from the water before they break down into nitrogenous waste. Protein skimming is the only form of filtration that physically removes organic compounds before they begin to decompose, lightening the load on the biological filter and improving the water's redox potential.

### Design

All skimmers have key features in common:

1. The skimmer body, where most of the contact between the Organic compounds and water takes place.
2. The foam separation area, or riser tube, where the organic foam is separated from the water.
3. A collection cup, where the foam spills over the riser tube and is drained away.



Source: [www.reefkeeping.com](http://www.reefkeeping.com)

Figure 2.4.4.2: Working principle of a Protein-skimmer.

Water flows through a chamber and is brought into contact with a column of fine bubbles. The bubbles collect proteins and other substances and carry them to the top of the device where the foam, but not the water, collects in a cup. Here the foam condenses to a liquid, which can be easily removed from the system. The material that collects in the cup can range from pale greenish-yellow, watery liquid to a thick black tar.

### *Principle*

Protein skimming removes certain organic compounds, by using the polarity of the protein itself. Dissolved Organic Compounds are bipolar molecules; these molecules can be described as hydrophobic (such as fats or oils) or hydrophilic (such as salt, sugar, ammonia, most amino acids, and most inorganic compounds) and are attracted to air/water interfaces, i.e. bubbles. A bipolar molecule contains one or more atoms attracted to air, and one or more atoms attracted to water. A skimmer exploits this difference in the following manner:

An air bubble moves through the column of organic-laden water, the electrically charged protein molecules (which contain electrically polar and electrically nonpolar regions) are attracted to the air/water interface of the bubble. The polar regions of the molecule (made up of nitrogen, oxygen, etc.) are attracted to the air/water interface and these polar side stick out away from the air bubble into the water column. The nonpolar regions stick out into the air bubble because it is repelled by the polar solvent (i.e. water).

### *Function*

A Protein skimmer works by mixing air with water to create tiny bubbles. Dirt, sludge and organic compounds stick to these bubbles. The smaller the bubbles, the more effective the protein skimming is. The surface area of small bubbles occupying the same volume is much greater than the same volume of larger bubbles. Large numbers of small bubbles present an enormous air/water interface for hydrophobic organic molecules and amphipathic organic molecules to collect on the bubble surface (the air/water interface). Once the dirt sticks to them, the bubbles begin to float up. When the bubbles reach the surface, they burst and deposit their collection of waste matter into the collection cup. The collection cup prevents the dirty deposit from slipping back into the column of water. The nature of salt water helps protein skimming, and because of this very reason, protein skimming is not feasible in freshwater [Proteinskimmer Helgeland, Brochure].

1. For optimum skimming, water flow thru the skimmer should be sufficiently slow as to allow interactions of an air bubble and organic waste. The best designs for

this are skimmers that employ water moving against the flow of bubbles. These are called counter-current skimmers.

2. The reaction chamber should be as tall as possible to maximize the contact time that the water has with the air in the skimmer, pumping as much air into the reaction chamber as possible.

3. The larger the amount of water to skim, the wider the diameter of the skimmer should be.

## 2.5 Diseases and Mortality

Halibut appear to have a non-specific immune system which becomes highly competent once they reach a size of > 2g. Up to a weight of about 10g, the young fish are still very vulnerable to diseases like *Vibrio* and IPN (infectious pancreatic necrosis), both of which also affect salmon, have caused particular problems [Pittman, unpubl.]. According to the Veterinary institute in Halibut are the main findings of atypical furunculosis and infection with various *Vibrio* species. But still bacterial infections are the biggest disease problems of marine fish [Hege Hellberg, Veterinærinstituttet Bergen].

Bacteria that develop in recirculation systems are typical of the marine environment. The highest concentration of fixed bacteria is found on the biological filter, which is the main source of bacteria in the culture system. *Vibrio*-type bacteria, some of which can be pathogenic, are present in very low numbers in the biological filter (as in any marine environment) and are destroyed by UV radiation before the rearing tank, although they can proliferate when the biological filter is overloaded with organic matter[[www.aquaflow.org](http://www.aquaflow.org)].

### A) Bacterial diseases

#### *Bacteria during egg phase and spawning*

As in other marine fish, the early life stages of the halibut are susceptible to opportunistic bacteria. The mucosal surfaces of eggs and larvae are colonised by bacteria, some of which are pathogens. Bacteria are found in large numbers on the surface of halibut eggs. This epiflora seems to be dominated by members of the *Cytophaga/Flavobacterium/Flexibacter* group, whereas *Vibrio spp.* are less frequent [Bergh-Kveite i opptrett]. The psychrophilic bacterium *Flexibacter ovolyticus* was isolated from halibut eggs that had suffered high mortality rates [Bergh et al. 2001]. Challenge experiments confirmed that this bacterium is

capable of causing mortality in halibut eggs and yolk-sac larvae by penetrating the eggshell [Bergh et al. 2001].

Challenge experiments with *V. anguillarum* on weaned halibut fry confirm that this bacterium is also pathogenic to later life stages of halibut.

The bacteria are visible as spots on the egg surface. These spots are easily visible in the lens and sometimes even with the naked eye. It is recognizable as long flexible rod bacteria with a creamy yellow pigmentation.

*Leucothrix mucor* is not very common on halibut and can easily be mistaken for fungi but can cause problems during hatching.

It is seen as partly long threads, which grow in rosettes. Rosette seats can be observed under the microscope at 400 magnifications [Bergh et al. 2001].

It has been speculated that strong growth of bacteria can cause oxygen deficiency in eggs, but it is unlikely that this is the case with halibut.

#### *Bacteria on larvae and juveniles*

At weaning and early nursery phase there have been incidents of serious disease outbreaks which have led to massive mortalities. This has been particularly prevalent in Norway, where the use of wild plankton (perhaps!) has led to the introduction of a variety of diseases.

The composition of the intestinal bacterial flora associated with yolk-sac larvae resembles the egg epiflora, whereas a shift in the intestinal microflora from a generally non-fermentative towards a fermentative flora dominated by the *Vibrio/Aeromonas* group coincides with the onset of exogenous feeding [Bergh et al. 2001]

Several species of *Vibrio* cause mortality in yolk-sac larvae. *Vibrio anguillarum* is transported across the intestinal wall and transported in the bloodstream to various organs and can cause bacterial infection of the bloodstream.

*Vibrio* bacteria are distinct so-called opportunistic pathogen, which means that they can survive in other ways than to parasite fish, but if the fish is weakened, they quickly will be able to cause infections that give rise to high mortalities.

Typical *A. salmonicida* subsp. *salmonicida* administered to yolk-sac larvae in a challenge experiment had more complex results. Although there was significant mortality, this probably resulted from the production of extracellular toxins produced by the bacterium, as histological and immunostaining examinations of the larvae revealed no evidence of bacteria in affected tissues [Bergh et al. 2001].

### *On-growing stage*

Comparing the pathogenicity of one atypical and one typical strain of *Aeromonas salmonicida* to subadult halibut (weight range 154 to 254 g) found no mortality when a minimum lethal dose of bacterium (typical *A. salmonicida* and atypical *A. salmonicida*) was injected into halibuts. A stress test of the survivors following the challenge showed that while 9 of the 87 halibut died, all were culture negative for *A. salmonicida*, suggesting that no carrier state had been established.

### **B) Virus Diseases**

Two viral diseases are known from Atlantic halibut in Norway. These are Infectious pancreatic necrosis virus (IPNV) and Nodavirus, giving rise to the disease VER (Viral encephalopathy and Retionpati)

#### ***IPN***

IPNV infections in halibut are so far registered in Norway and Scotland. Mortality in fish may exceed 90%. Sick fish shows rotation around the longitudinal axis. White threads of faeces and tarmvev can hang from gone. In the liver, kidney and spleen necrosis can be seen on histological samples, and one can see the rejection of tarmvev. In infection experiments have seen a tendency for susceptibility to infection increases with higher temperature and decreases with increasing fish size.

#### ***Nodavirus infections***

Nodavirus infections have given rise to significant mortality in several commercial production facilities for halibut in Norway, at least from 1995.

Nodavirus attacks the central nervous system and causes damage to nerve tissue, mainly retinal, brain and spinal cord. Such viruses have so far described from 22 different marine bony fish species over large parts of the world.

Since the central nervous system is damaged, it gives rise to significant behavioral changes. Halibut larvae stop eating and become very lean. Abnormal swimming behavior like rotation around the longitudinal axis and circular movements are common. Dying halibut are often lying on the blind side up.

In infection experiments with the yolk sac larvae of halibut, the virus was first detected in the medulla oblongata, and then spread back into the spinal cord and forwards to the brain. The retina was the last body that was affected. Nodavirus detected in cells outside the nerve tissue. It is not known what factors affect the









virus's ability to infect nerve cells. Findings suggest that target cells must reach a certain stage of development before the infection is possible. This stage is reached during the yolk sac stage, and mortality occurs in the fourth week after hatching at 6 ° C. Observations of Nodavirus infections on halibut farming is essentially related to the first feeding phase, but it is possible that the infection can take place already in the yolk sac phase. Transmission experiments show that if the virus is introduced in sufficient quantities rapid spread and high mortality must expected.

Studies are underlining the need for maternal vaccination and early vaccination during the live food stages. It is therefore tempting to speculate that the major transmission route of Nodavirus is from broodstock, where it can hide in a subclinical state until some mechanism activates it at the time of maturation and spawning, when it follows eggs or sperm to the offspring. If a few of the offspring in a rearing unit acquire the disease, the virus will be released into the environment, initiating horizontal transmission with high mortality. Some of the larvae/fry, however, will survive, but may still harbor the virus, which could then be transmitted to the next generation. But, as the virus occurs at such high concentrations in the affected units, another possibility is that it can persist in the environment from one production batch of larvae to the next. As Nodavirus is very stable and can survive in seawater for a long time further research should concentrate on the measures that have to be implemented to eliminate the virus once it has entered a sea-farm [Nerland et al. 2007].

The factor that control the propagation of the virus as to why the virus suddenly breaks out in a group but not in another is unknown.

### ***Other***

It is very likely that there will be several viral diseases in farmed halibut than the two that are known so far. Two current virus groups; *Rhabdovirus* and *Herpes viruses*, which are both known from flatfish has been reported from turbot in Scotland and Ireland, and a similar virus has been reported from the Japanese flat fish Hiram.

| ORGANISM                           | IMAGE   | COMMENT  |
|------------------------------------|---|--|
| <b>PARASITES</b><br>Trichodina sp. |    | Not uncommon with halibut<br>Treat with Formalin<br>Not a serious threat if controlled                                       |
| Tapeworms                          |    | Status unclear with halibut<br>Probably not a serious threat   |
| Entobdela sp.                      |    | Monogenean skin parasite<br>Treatment techniques currently being developed. Can be heavy infestations                        |
| <b>BACTERIA</b><br>Aeromonas sp    |    | Furunculosis-type problems<br>Could potentially affect halibut, and can probably act as carriers<br>Vaccination, Antibiotics |
| Vibrio sp                          |   | Can certainly affect big halibut, but probably on if weakened<br>Vaccination, Antibiotics, Good Husbandry & Nutrition        |
| <b>VIRUS</b><br>IPNV               |  | Can certainly affect <2 g halibut and 20- 100g halibut. Probably can also act as carriers.<br>Avoid challenge                |
| VHSV                               |  | Halibut seem to be v. resistant to this virus - but a new strain could cause problems. Avoid risks (eg no wet fish feeds)    |
| Nodavirus                          |  | A problem in Norway. Vertical transmission.<br>Cull suspect broodfish  |

Source: Seafish industry

Figure 2.5: Overview over possible pathogens affecting Atlantic Halibut.

### 2.5.1 Vaccination and Non specific Immunstimulation

Lymphoid organs, i.e. the thymus and kidney, develop, and immunoglobulin-bearing cells proliferate, around the time of first feeding, whereas the spleen could not be found before exogenous feeding. Vaccination before this stage may therefore be impossible. However, there are indications that weaned halibut at a size of 0.1 g can be vaccinated against vibriosis [Bergh et al. 2001].

Stimulation of the non-specific immune system is an alternative method, which could be suitable for earlier ontogenetic stages. Macrophage stimulator FMI and the immunomodulator, laminaran ( $\beta$  (1, 3)-glucan) has been tested in experiments on halibut yolk-sac larvae, however with no results.



The search for an effective immune-stimulant, which would be more resistant to bacterial degradation, led Dalmo et al. (1998, 2000) to the bacterial lipopolysaccharide (LPS) obtained from cultures of *Aeromonas salmonicida subsp. salmonicida*. LPS is an interesting candidate as an immunostimulant, since it is an important constituent of vaccines based on killed bacteria, and has been shown to activate fish macrophages and B-cells *in vitro* and *in vivo* after injection. LPS was taken up by the larvae and survival was significantly improved in comparison with untreated.

Recirculated water may give conditions similar to the microbial matured water system approach for rearing marine fish larvae. The more stable physical environmental conditions in the recycled water groups may also be conducive to better rearing performance.

## 2.5.2 Pharmacokinetics

### *Antibiotic use in Aquaculture*

Antibiotics are drugs of natural or synthetic origin that have the capacity to kill or inhibit the growth of micro-organisms. In aquaculture, antibiotics have been used mainly for therapeutic purposes and as prophylactic agents. The feeding of antibiotics is associated with decreases in animal gut mass, increased intestinal absorption of nutrients and energy sparing. Antibiotics may enhance the uptake of nutrients from the intestine by thinning of the mucosal layer and by inhibiting intestinal bacteria, which inactivate pancreatic enzymes and thereby decrease the digestibility of dietary protein [Pilar Hernández Serrano, 2005]. Also there is indirect evidence that survival of yolk-sac larvae in small-scale static systems requires the addition of antibiotics due to bacteria causing mortality in commercial halibut larval rearing systems [Verner-Jeffreys et al. 2003].

In other experiments the addition of antibiotics caused a short-term reduction in the total numbers of bacteria in the tank water. However, yolk-sac larvae remained practically free of culturable bacteria for a much longer period, despite the high concentration of microorganisms in the tank water. This could be due to accumulation of antibiotics within the larvae, prolonging their resistance to later colonisation by bacteria.

At present, 6 antibacterial agents are used in aquaculture in Norway; the quinolones Flumequine and Oxolinic acid, the Tetracycline derivative oxytetracycline, the potentiated Sulphonamide Tribissen (trimetoprim + sulphadiazine) and Florfenicol, which is an Amphenicol derivative. Antibacterial agents are usually administered as medicated food pellets, with the antibacterial either coated on the surface of the pellets using a small quantity of oil or incorporated into the pellet.

But because bacteria multiply quickly and also have the ability to adjust to different environments, feeding antibiotic lead to antimicrobial resistance and these resistant bacteria can transfer the resistance to other bacteria (even to bacteria of different genera) that have never been exposed to the antibiotic, and this phenomenon is known as horizontal gene transfer. In fish farming (aquaculture, mariculture, etc.), this has led to the development of antibiotic resistance in *Aeromonas hydrophila*, *A. salmonicida*, *Edwardsiella tarda*, *E. ictturali*, *Vibrio anguillarum*, *V. salmonicida*, *Pasteurella piscida* and *Yersinia ruckeri* [Verner-Jeffreys et al. 2003 and Naviner et al. 2006].

## 2.6 Environmental issues

### 2.6.1 Persistent organic pollutants

Liphophilic persistent organic pollutants (POPs), such as polychlorinated biphenyls (PCBs), organochlorine pesticides (DDTs) and hexachlorobenzene (HCB), are oily liquids or solids, clear to yellow in color, with no smell or taste. PCBs are very stable mixtures that are resistant to extreme temperature and pressure. They were used in hydraulic fluids, heat transfer fluids, lubricants, and plasticizers.

Commercial production of PCBs ended in 1977 because of health effects associated with exposure [Illinois Department of public health, Feb.2009].

Historically, most of the fish in Europe's Rhine River were killed by the discharge of pesticides. Some pesticides, such as pyrethroid insecticides, are extremely toxic to most aquatic organisms. It is evident that pesticides cause major losses in global fish production. Up to 70% of all organochlorinated compounds entering the environment are concentrated in the world's oceans [Sterlli 2009].

#### *Origin*

These chemicals come directly from rivers or the atmosphere into the sea as results of agricultural and wastewater contamination through fallout, drainage, or runoff erosion, and from the discharge of industrial and shipping effluents.

Once in the environment, PCBs can be transported long distances and they bind strongly to soil and sediment so they tend to be persistent in the environment.

They have been found in air, water, soil, and sediments throughout the world [Illinois Department of public health, Feb. 2009].

The extent to which a pesticide runs off an agricultural field is determined by the unique combination of climatic, soil, and management factors that characterize each field, crop, and year combination [Altinok et al. 2005].

Currently there are approximately 1.2 million t of proven Polychlorinated biphenyls (PCBs), in the environment (ocean, atmosphere, groundwater and land) detectable.

Approximately 100,000 t were disposed in the North Atlantic, for which it is the world's most contaminated water with PCBs [blog.oceancare.org/2009/08].

Different researches have continued to reveal that contamination in deep-sea biota is more elevated than surface living species, highlighting transport processes of POPs to the deep-sea environments.

For example, Froescheis et al. 2000, showed that the PCB levels in bottom dwellers at depths greater than 800 m were between 10 and 17 times higher than in the related surface species.

### *Effects on fish*

According to different researches contaminants can act in different ways; as signals, modify odorant perception, act on the nervous system and/or other physiologic responses, all of which potentially alter normal olfactory mediated responses.

While exposure to these pesticide concentrations may not necessarily produce toxicity, the comparison to olfaction shows that these concentrations might be capable of producing a biological response.

Chemical contaminants in the aquatic environment have been implicated in disrupting the chemosensory abilities of fish, as well as its documented oestrogenic effects.

Similarly, many pesticides are implicated in a loss of receptor function in fish [Tierney et al. 2007]. The presence of pesticides, e.g. DDT and PCB, in the aquatic environment has been associated with many diseases, including 'cauliflower disease', lymphocystis and ulceration and liver neoplasia [Moore and Lower 2001]. Malformations in common dab, flounder, plaice (*Pleuronectes plutessu*) from the southern North Sea during 1984-1995 were considered to be linked to pollution with organochlorines. Thus as a result of long-term surveys, these authors considered that the malformations resulted possibly from low water temperatures that predisposed the embryos to the effects of organochlorines.

Liver disease, including neoplasia, has been described in winter flounder (*Pleuronectes americanus*) from Boston, USA, particularly in the region of a sewage outfall [Moore and Waring 1996]. Of relevance, these workers noted that during 1987-1993, there was a reduction in the incidence of neoplasia concomitant with a decline in output of chemicals, notably DDT and other chlorinated hydrocarbons, into the receiving waters.

In the real world, aquatic environments may often be contaminated simultaneously by more than one, and often several, different chemicals. The possibility exists therefore that different contaminants, each disrupting a separate element of the

chemical communication pathway, may act in concert to seriously impair or to knock out the chemosensory abilities of fish at levels below those currently considered to be a problem based on single-chemical ecotoxicological studies.

An example is tributyltin (TBT), which was used in underwater paints for ships - preventing fouling of ship hulls by algae, barnacles and mussels. TBT acts like a hormone on marine snails and affects the reproductive organs of the worm so that the animals become infertile. Infertile screw can now be found worldwide, especially in ports and sea routes, over 100 species of marine snails are threatened with extinction.

### **Heavy metals**

There are 35 metals special interest because of occupational or residential exposure; 23 of these are the heavy elements or "heavy metals": antimony, arsenic, bismuth, cadmium, cerium, chromium, cobalt, copper, gallium, gold, iron, lead, manganese, mercury, nickel, platinum, silver, tellurium, thallium, tin, uranium, vanadium, and zinc. Small amounts of these elements are common in our environment and diet and are actually necessary for good health, but large amounts of any of them may cause acute or chronic toxicity [www.lef.org]. The relationships among tissue metal burdens and overall toxic response during chronic exposure is complicated by the fact that metals accumulate differentially in tissues. An additional complication is that some metals, for example Cu and Zn are essential minerals with cellular and tissue levels subject to metabolic regulation [McGeer et al. 1999].

### *Origin*

Heavy metal pollution can arise from many sources;

### **Lead**

Lead is a very soft metal and was used in pipes, drains, and soldering materials for many years. Millions of homes built before 1940 still contain lead (i.e. in painted surfaces), leading to chronic exposure from weathering, flaking, chalking, and dust. Every year, industry produces about 2.5 million tons of lead throughout the world. Most of this lead is used for batteries. The remainder is used for cable coverings, plumbing, ammunition, and fuel additives. Other uses are as paint pigments and in PVC plastics, x-ray shielding, crystal glass production, and pesticides. Target organs are the bones, brain, blood, kidneys, and thyroid gland [Roberts 1999; ATSDR ToxFAQs for lead].

## **Cadmium**

Cadmium is a byproduct of the mining and smelting of lead and zinc. It is used in nickel-cadmium batteries, PVC plastics, and paint pigments. It can be found in soils because insecticides, fungicides, sludge, and commercial fertilizers that use cadmium are used in agriculture. Cadmium may be found in reservoirs containing shellfish. Cigarettes also contain cadmium. Lesser-known sources of exposure are dental alloys, electroplating, motor oil, and exhaust. Target organs are the liver, placenta, kidneys, lungs, brain, and bones [Roberts 1999; ATSDR ToxFAQs for Cadmium].

## **Aluminum**

Although aluminum is not a heavy metal, it is readily available through the use of food additives, antacids, buffered aspirin, astringents, nasal sprays, and antiperspirants; from drinking water; from automobile exhaust and tobacco smoke; and from using aluminum foil, aluminum cookware, cans, ceramics, and fireworks [ATSDR ToxFAQs for Aluminum].

## **Copper**

It can be leached from pipes, leaving in severe cases a greenish ring on bathroom fixtures. Water coolers and ice-makers in refrigerators also use copper tubing. Water that sits in these units can contain dangerously high levels of copper.

## **Copper toxification**

At the organ level, fish resemble higher vertebrates with respect to copper homeostasis. Copper is an essential element that serves as a cofactor in a number of enzyme systems for most living organisms but, at high concentration, copper become a toxic pollutant [Carvalho et al. 2004]. Copper concentrations in water can lead to increased copper levels, particularly in the gills and the liver [Grosell and Wood 2002]. Research about copper accumulation in the liver of copper exposed fish was greater at lower temperature and pH 8.0. Copper accumulation is related to copper toxicity and the water pH is a determinant factor in this process [Carvalho et al. 2004].

### **2.6.1.1 Effects on fish**

- Reduced appetite [McGeer et al.1999]
- Reduced immune response; when copper is too high in the body an imbalance between copper and zinc develops, leading to all kinds of infections [Dr. Wilson, 2011].
- Affecting the nervous system; Copper stimulates the production of Neurotransmitters, an imbalance can lead to abnormal behaviours like gasping for air at the water surface and apathy, which is a kind of depression [Dr. Wilson, 2011].

Higher copper levels increase internal mobilization of metal binding proteins such as metallothionein, which are assumed to act as detoxification and storage mechanisms. However, fish show fast physiological recovery from copper effects and long-term gill histological recovery after transference to water free of copper [Carvalho et al. 2004 and McGeer 2000].

## **2.6 Behavior**

### *Natural*

Halibut's behavior has for thousands of years evolved to be the best in survival and reproduction in the natural surroundings. Before halibut become big enough to be inaccessible for most predators, there will be a fight between eating and fast grow in order to avoid exposure to the risk of being eaten themselves. Camouflage and low swimming activity are the two main method of defense.

Halibut continues to undermine movements even if there is no sand in the tanks, and they still have a strong instinctive need to camouflage for predators even though there are none in the tanks.

### *Aquaculture*

#### **Bottom behavior**

More than 80 % of the time halibut lay quiet on the bottom. The rest position is both head and tail down toward the bottom, and halibut have camouflage pattern similar environment. When the halibut will settle down in a place often performs the digging motion with their body to dig into the sediment and stir up sand from the bottom. When halibut land on one another on the bottom, they are apparently little disturbed and will usually remain motionless in the same position. If the other halibut will remain for a while, still move a bit to get in a better position, but if there is too much interference, will one leave the bottom and find itself another

place. With increasing densities, there will be more disturbances and swimming activity per individual will increase, leading to the total rise in activity in the tank. It will be much movement along the bottom and a lot of landing and setting off from the bottom.

Halibuts could be completely immobile for periods of 5-10 hours, usually around midnight, which indicates that halibuts need a long "sleep" or rest periods.

Dissatisfaction in tanks with high density may be due to increasing disturbance and deprived of these periods, which may cause "sleep deprivation" and low feeding motivation.

### ***Eating Behavior***

Halibut mainly use vision to detect prey, and can take both movable and immovable prey. By feeding flowing feed they prefer to catch the pellet before it rises to the surface. Also, halibut that has lost one eye has more difficulty to take pellets in the water column. In comparisons between flowing feed and sinking feed we have not found any growth differences.

### ***Aggression***

The halibut is the most aggressive being a fry and small halibut stadium (5-150 g), but bites and aggressive actions observed also in larger halibut. Large individuals grow different within a group especially when fish growth is due to genetic differences and/or early establishment of stable size ranks (hierarchies).

These behavioral interactions can lead to decreased growth rate in low ranking individuals. Large differences in individual growth rates have been noted in juvenile halibut [Haug 1990], as have stable size hierarchies and aggressive behavior [Jonassen et al. 1999].

A big problem with aggression is eye damage. Fish that are bitten get eye infections that often lead to the loss of the eye. This leads further to reduced weight gain in the period during the wound has healed and the fish will later have more problems with taking food. The wound is usually healed after a few months time, and halibuts seem to do well with one eye and grow just as well as normal fish after the wound has healed. Fish losing both eyes become blind/dark (cannot regulate the camouflage pattern) and growth is poor, but can still survive for a long time. This behavior seems to be stress induced, as one often gets the increasing frequency of eye injuries when, for example, the temperature varies a lot or some kind of additional stress. It is therefore important that the environment is most stable and that the temperature of the water intake is not affected by the weather. Also the quick change from light to dark can lead to "panic" and stress.



Sorting of the fish in groups of uniform size reduces the eye picking problem. The use of larger vessels also has a positive effect.

In small fry it has been observed that the aggressive fish attack the individual fish or groups of 2-3 fish that is alone on the bottom. In a study in Scotland, they were significantly less damage and better growth when the density increased from 500 juveniles (3-6 cm) per m<sup>2</sup> to 2000 fry per m<sup>2</sup>. Bite injuries were also more severe in low-density and consisted of wounds on his back, tail and anal fins. There was no eye damage or injury to the pectoral fins in this experiment.

### ***Abnormal behavior***

In halibut farming there are different forms of abnormal behavior, which naturally halibuts would not do. A typical example is surface swimming or “dipping”, where the fish swims almost vertically in the surface and with heads of and partly above the surface. In experiments where fish have been marked with PIT tags has shown that most fish with this behavior have on average much lower feed intake and poorer growth than those that swim little, too much surface swimming is a bad sign for the farmer. Video recording shows that halibuts swim straight up from the bottom and begins to nod, so this is a deliberate act. One hypothesis suggests that this is a form of stress management and self-stimulation, wandering back and forth like a tiger in a cage. This activity decreases if the fish get larger and with deeper vessels and we can also interpret this behavior as a sign that the fish wants to get away from a situation it cannot cope with.

In juveniles swimming in a spiral, or spin around and around on the surface can be a sign of viral infection, but also an abnormal behavior caused by unsatisfied needs in the environment. By moving such fish to other tanks they usually calm down.

### **2.6.1 Stress**

Almost all environmental factors tested, can influence the degree to which fish respond to stressors. External factors include acclimation temperature salinity, time of day, wave length of light and even background color of the tanks [Barton 2002]. But also the genetic component plays a role in stress responses, according to Barton some fish may be predisposed to consistently release high or low stress hormones to stressors.

Environmental stressors have been grouped broadly as primary and secondary. Primary responses include a number of hormonal changes while secondary stress responses are related to physiological adjustments such as in metabolism,

respiration, acid-base status, hydro-mineral balance, immune function and cellular responses [Barton and Iwama 1991].

Environment, which does not meet the needs of the fish, will lead to chronic stress and frustration and fish individuals develop different strategies to cope with this situation. Some choose a reactive strategy (wait for it to go over) and become lethargic and lying quietly on the bottom and not worry about eating or other activities. Others will choose an active strategy with abnormal swimming activity to try to change the situation.

When fish are moved, the characteristics of the water should be matched as closely as possible to the water source. Stress can occur if changes in water temperature and quality are abrupt, even when values are within the tolerance range of the species; and definitely when they are outside of the range [Wedemeyer 1997]. Barton and Iwama 1991 tested different aquaculture species on releasing stress hormones after 2, 4 and 8 h of transport, which showed that the level of these hormones were up to 25 times higher than before the transport.

In most halibut groups are individuals who show remarkable ability to adapt to unusual environments, such as very high density, eating and growing well under difficult circumstances. In the future, a farmed halibut must be breed adapted to farming community so that a large proportion of halibuts master the situation [sintef.no].

## 2.7 Atlantic Halibut farming at AGA Halibut

### 2.8.1 History and Development

Aga Halibut Dønna was established in November 2004. Investors from Bømlo bought the bankrupt estate of Dønna Marine Holding from Helgeland Sparebank and Rana Savings Bank. Also because the Hatchery Aga Marine in Bømlo needed costumers for their juveniles, they decided to have their own on-growing. Aga Halibut in return bought Aga Marine in Bømlo.

Due to problems and changes within the company Aga Marin in Bømlo was closed down in 2010 and instead the Hatchery in Risør was bought.



Aga Halibut Risør Aga Halibut Dønna

## 2.8.2 Hatchery in Risør

### 2.8.2.1 Structure and Organization

#### Facilities

Risør has 15 egg incubators with 350 liters each and 14 larval silos with 6 m<sup>3</sup> each.

There are total of 18 startfeeding tanks distributed into 2 rooms: 15 tanks have diameter of 2, 5 m resulting into 5 m<sup>3</sup> volume each and 3 tanks with 3 m in diameter resulting in 7 m<sup>3</sup> each.

#### Water flow, Salinity and Temperatures

Table 2.8.2: Increasing water flow by days in the 5m<sup>3</sup> tanks.

| Startfeeding | Day | Water flow | Unit  |
|--------------|-----|------------|-------|
|              | 0   | 15         | l/min |
|              | 3   | 20         | l/min |
|              | 5   | 30         | l/min |
|              | 11  | 35         | l/min |
|              | 19  | 42         | l/min |

Source: Risør Fisk

Temperature in incubators and silos are 6°C +/-1 and 11°C +/-1 in startfeeding tanks, the salinity is 32 ‰ +/-1 ‰

### 2.8.2.2 Feeding and Development

The fry hatches 12-13 days after fertilization, are 10 days in the incubators and approximately 45 days in the silos.

During the 60 to 90 days startfeeding period the fry are fed short time with Artemia enriched on Multigain (sold by Biomar), and later with 5 days ongrown Artemia (also enriched on Multigain).

After the startfeeding the juveniles are weaned on Aglo Norse feed, and then after approximately 2 to 3 weeks gradually on Skretting feed.

## 2.8.3 On-growing site at Bjørn

### 2.8.3.1 Structure and Organisation

Aga Halibut has two recirculation systems and each provide for 20 fish tanks. One single tank has a volume of 251 m<sup>3</sup> and water flow rate of 0, 9 m<sup>3</sup>/minutes. Both systems have a UV-unit, Drumfilter, Biofilter and Degasser and in system A is additionally a Proteinskimmer with an ozone generator installed. The water gets

filtered up to 98%, with new incoming water only at 2%. The inlet pipe is ca 1000 m long and reaches 90 m deep into the Asværfjord. There are two main pumps serving the systems with fresh water and via pipe (PE) the water reaches into the puffer tanks, there it is stored and c.a. 2% of it is channeled into the recirculation systems.

At times a flow- through system was installed, obtaining water directly from the sea. After the water was sucked through the main pumps it was further transported via a collective pipeline leading into the farm. Most of the water was diverted at the first joint of this pipeline, transporting the water to the flow-through tanks. In all system, recirculation and flow- through, the water was oxygenated. Two tanks usually shared an oxygen cone, which received oxygen from the main battery. This battery was regularly filled up with compressed oxygen, delivered from Adox - Oxystream. Oxygen was initiated into the inlet pipes of each tank, where it mingles with the water.

Figure 2.8.3.4 gives a simple overview over the water flow. The water which comes from the fish tanks goes through the drumfilter and 30 % of that runs through the UV- unit. All the water further goes through the degasser and is being collected in the header tank. A partial water flow, which is harbored in the drumfilter runs together with water from the fish tanks into the biofilter. The biofilter has its own internal loop in the water treatment.

The Proteinskimmer gets water from the header tank and leads it further to the degasser.

#### *Proteinskimmer with ozone generator (Figure 2.8.3.1)*

The type of Proteinskimmer is called Helgoland and manufactured by ENVICON. The water flow rate is 6, 3 m<sup>3</sup>/minutes and it cleans approximately 40 % of the recirculation water. It is connected to an ozone generator, which enhances the effect of the Proteinskimmer and is further connected to the suction well (header tank) and degasser. The ozone generator produces 200 g ozone per hour, which corresponds to 100 % capacity. But mostly only 90 % of its capacity was used, so approximately 180 g of ozone per hour were produced.



Foto: Lydia Michalski

Figure 2.8.3.1: Proteinskimmer at Bjørn.

### Drumfilter (Figure 2.8.3.2)

Was manufactured by Cimbria Aquatech and was equipped with a filter opening of 50  $\mu\text{m}$ . It has a water flow rate of 27  $\text{m}^3/\text{minutes}$ . It receives water from the fish tanks and is further connected to Biofilter and the Degasser.



Foto: Lydia Michalski

Figure 2.8.3.2: Drumfilter at Bjørn.

Biofilter (Figure 2.8.3.3)

Was manufactured by Cimbria Aquatech and is categorized as a low pressure system filled with bioblocks, which have the task to increase the filter area. The water flow rate 10 m<sup>3</sup>/min and it cleans 450 kg Feed per day maximum. It follows after the drumfilter and is connected to the suction well (header tank).



Foto: Lydia Michalski

Figure 2.8.3.3: Biofilter at Bjørn.

UV

It has 30 Lamps and power of 65 Watt disinfecting 30% of the recirculated water. It receives water from the drumfilter and is connected to the degasser.

Degasser

The degasser was manufactured by Cimbria Aquatech and is a cascade air-filter which is filled with bioblocks where the water runs through. It has a water flow rate of 30 m<sup>3</sup>/min receiving water from the drum filter and the UV disinfection unit and is connected further to the suction well, which mingles the new incoming- with the recirculated water. The suction well distributes the all water to the fish tanks.

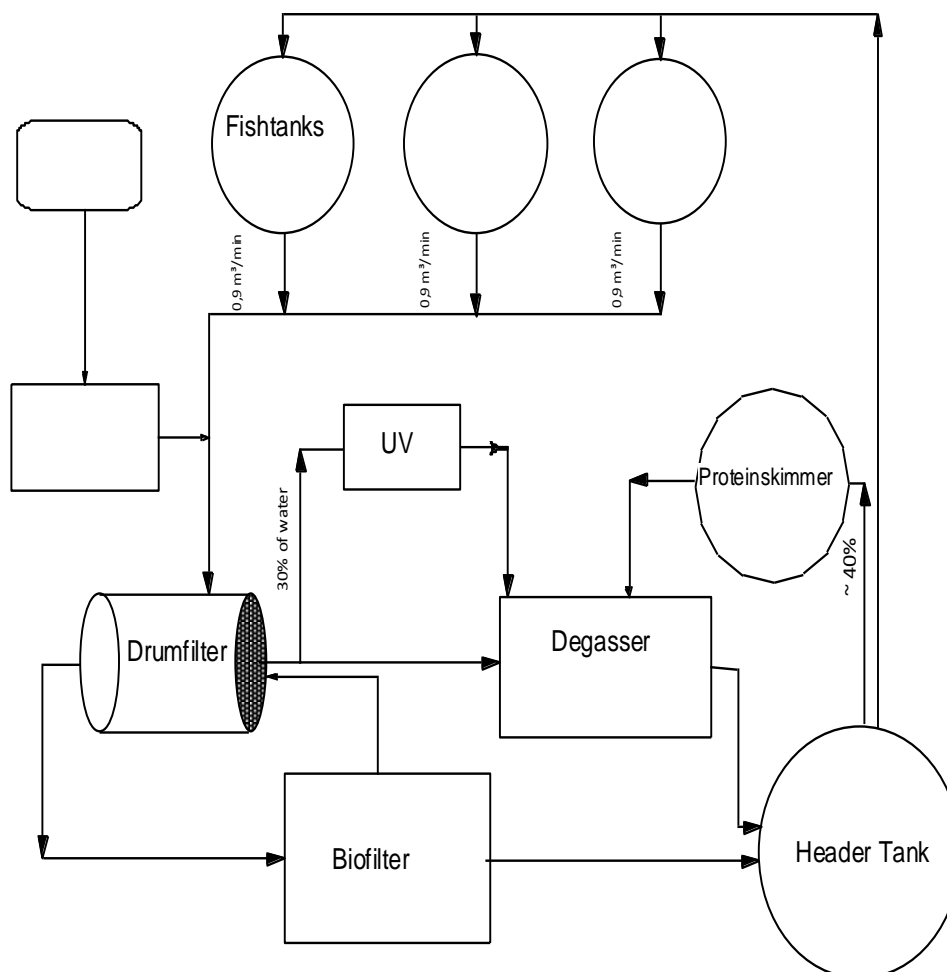


Figure 2.8.3.4 Simple water flow chart of the recirculation system at Bjørn.

### 2.8.3.2 Feed and feeding technique

The feed of use is Amber Neptun ST for small “Settefisk” (10-40 g) and Europa Marine S for large fish provided by Skretting. The Ingredients are fish meals, pre-digested fish meal, Prawn/shrimp meal, fish oil, wheat, and maize gluten meal, Betaine, Vitamins and Minerals [Skretting].

Over each tank is either a rotating or bell feed distributor (Figure 2.8.3.4), which is regulated by a PC-controlled feeding system.

6 Silos (Figure 2.8.3.4, right) with a maximum capacity of 1, 36 t of feed are connected over pipes to the feed distributor and via air pressure reaches the feed to the tanks.





Foto: Lydia Michalski

Figure 2.8.3.5: Bell Feeder (left) and Silos (right) at Bjørn.

#### 2.8.4 Mortality issue among newly arrived Halibut at Bjørn

Aga Halibut has been taken in on-growing halibut since 2003/2004. The fish origin from various hatcheries in Norway and Island are delivered via truck. The transport usually takes 24 to 48 h and the fish are hold in batches with water temperatures of 4 to 6 °C. The oxygen and water temperature are monitored throughout the transport.

After the fish are set into tanks they seem to be normal; laying down at the bottom and eating. The picture changes exactly after 6 week; many fish start to show abnormal behaviour, non eaten feed at the tank bottom and a rise in mortality.

##### *Abnormal Behaviour*

The so called “dupping”, where the fish snap for air on the water surface, constant swimming as well as being lethargic are the significant behavioural abnormalities, observed.

##### *Mortality development*

The mortality starts around week 6 after arrival and has its maximum for about 2 weeks and slowly decreases after that. The amount of dead fish varies also between fish groups and definitely fish size plays a role. Table 2.8.4.1 shows the average mortalities that occurred at Bjørn in the years 2004 until 2009.

Table 2.8.4.1: Average mortalities per month.

| Size [g]        | 0-100 | 50-150 | 150 -500 | 500-1000 | 1000-1500 |
|-----------------|-------|--------|----------|----------|-----------|
| Mort./month [%] | 7,01  | 5,45   | 1,07     | *4,8     | 0,6       |

\*only one fish group with this size was listed at Farmkontroll

But mortality occurrence differed a lot from group to group, while it was with some groups around 3 % per month was it with others 10 % per month with same size and same hatchery origin [from farmkontroll].

#### Attempts to solve the problem

1. Veterinary examination for Bacteria, Viruses other diseases
  2. Control of water parameter, gas pressure and metals
  3. Taking in different fish sizes; from 2 g to >100 g
  4. Flow -through system
- 
1. Veterinary examinations have shown that the fish do not suffer from bacteria such as Trichodina, Costia, atypical Aeromonas or Atypical furunkulosis. Though these have been found randomly and in low amounts so at least they cannot be considered as a primary cause for the mortalities. IPN and Nodavirus has been tested negative also and no know parasite has been found either.

#### The diagnoses however that were made repeatedly over the years:

- Eye and fin injuries, mostly caused by mechanical impact.
  - Liver proliferation and hypertrophie
  - Liver cirrhosis
  - Gill inflammation and hypertrophie
- 
2. Water quality parameters were tested weekly for NH<sub>3</sub>, NH<sub>4</sub>, Nitrate, Nitrite, CO<sub>2</sub> and pH and the values were within the normal range. Also the total gas pressure (TGP) has been tested normal.

2007 was the intake- and recirculation water in the farm tested for heavy metals. Lead and Copper were categorised as high occurrence and also were the values higher than in the intake water (all metals see appendix table). For the other metals the values were in low range and therefore not harmful to the fish.

Table 2.8.4.2: Kopi of the heavy metal results analysed from the incoming water and the water in the recirculation system at Bjørn, 2007.

| Parameter     | Unit | Intake water | Recirculating water |
|---------------|------|--------------|---------------------|
| Arsen         | µg/l | 1,2          | 1,8                 |
| <b>Lead</b>   | µg/l | <b>0,6</b>   | <b>1,1</b>          |
| Cadmium       | µg/l | < 0,05*      | 0,05                |
| <b>Copper</b> | µg/l | < 0,5*       | <b>2,2</b>          |
| Crom          | µg/l | 0,6*         | 1,3                 |
| Mercury       | µg/l | 0,002        | 0,002               |
| Nickel        | µg/l | < 0,5        | 1,5                 |
| <b>Zink</b>   | µg/l | < 4*         | <b>58</b>           |
| Silver        | µg/l | 1,5          | < 0,5*              |

Source: Molab AS, 8607 Mo i Rana , 12.3.2007.

\*Elements were not detected in the samples but the detection limit for the analysing method was higher than the given condition classes.

In this analyses lead, copper and zinc were found in higher and strongly polluted amounts, so that the plant was searched for metal- water contacts. It was found that several zinc anodes were used in the system and the propellers in the 2 main pumps were made out of bronze, which consists to 80 - 90 % out of copper. The propellers were therefore coated with a ceramic layer and the zinc anodes were removed except the ones in the drumfilter. But after these changes had been done, no security test has been made.

3. Another attempt was to put in fish with weight < 2 g, which was based on the idea that the immunsystem will adapt to the natural bacterial occurrence at Bjørn, while it develops. But the high mortality remained.
4. The mortality in the flow-through system ranged from 2, 1 - 24, 6 % per month with fish size from 50 to 150 g. A lower water temperature (7 to 8 °C) reduced the growth of the fish drastically and however the mortality development was similar to the ones in the recirculation system but the amount was lower.

### 3. Material and Method

#### 3.1 Experimental design and Technical setup

The experiment was carried out at the land based and sea cage site on Dønna in during 27.04.2011 until 17.08.2011 altogether 119 days.

The set up was based on the question if the proteinskimmer can improve the growth rate and lower the mortality plus if Antibiotic has an effect on the mortality extent of the fish. The corresponding Nullhypothesis =  $H_0: \mu_1 = \mu_2 = \mu_3$  against the alternative Hypothesis =  $H_1$ : At least one  $\mu_i$  is different from the other population means.

#### **The model for the mortality is:**

$Y_{ij} = \mu_i + e_{ij}$ . With  $\mu_i$  as distribution mean,  $Y_{ij}$  = Mortality rate of small ongrowing halibut  $i$ , observation number  $j$ ,  $i = 1, 2, 3$  and  $j = 1, \dots, n_i$  and  $e_{ij}$  is normal distributed  $[N(0, \sigma)]$ . All the error-terms are independent.

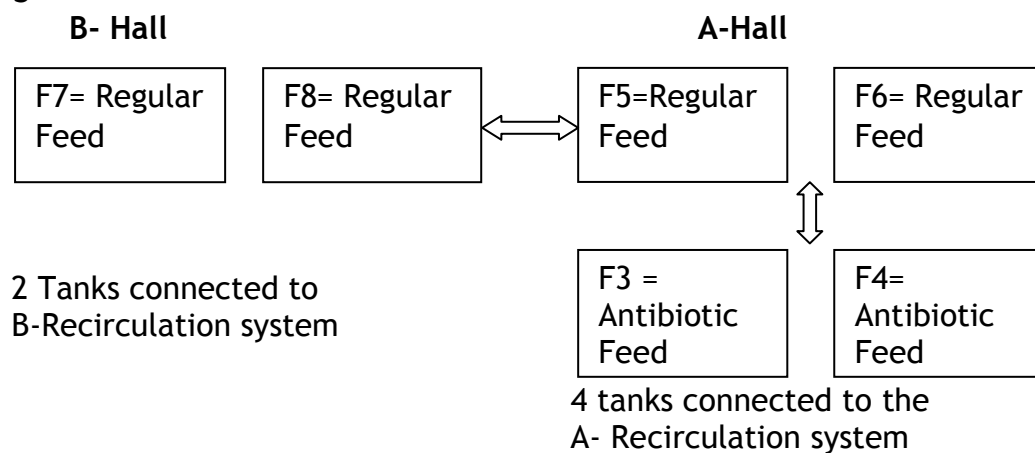
#### **The model for the growth rate is:**

$Y_{ij} = \mu_i + e_{ij}$ . With  $\mu_i$  as distribution mean,  $Y_{ij}$  = Growth rate of small ongrowing halibut  $i$ , observation number  $j$ ,  $i = 1, 2, 3$  and  $j = 1, \dots, n_i$  and  $e_{ij}$  is  $N(0, \sigma)$ . All the error-terms are independent.

In order to compare different water qualities, the effect of antibiotic feed as well as landbased - with sea cage farming, the design was set up as followed;

- 2 tanks were exposed to standardized feed and recirculated water,
- 2 tanks were exposed to standardized feed and recirculated water treated additionally with a Protein skimmer and Ozone.
- 2 tanks were exposed to antibiotic feed and recirculated water treated additionally with a Protein skimmer and Ozone.
- 2 sea cages, 2 x 2 m, set out directly into existing cages.

Design:



Sea cages:



The rectangular tanks have an external in- and a flat centralized outlet and carry a water volume of around 1440 l.

The water flow is between 34 to 45 l/min so the retention time stays between 43 to 23 minutes, creating a small self cleaning effect in the tanks though the bottom has to be cleaned every 2<sup>nd</sup> week additionally.

Pure oxygen is supplied via pipes for all 6 tanks and the amount is regulated according to the measured O<sub>2</sub> value in the outlet, which remained above 75 %/ 6, 6 mg/l at a temperature of 11 °C and a salinity of 35 ‰.

The amount of water and oxygen is regulated manually by valves and the tanks are flushed every day to prevent clogging of the outlet. Each tank has an automatic disc feeder.

### 3.2 Fish material and Rearing Conditions

Tanks

The main experiment, with fish from AGA`s own hatchery at Risør, the average weight was 22 g. 6000 was the amount of fish during experiment.

The fish were distributed into 6 recirculation tanks with 1000 to 1300 individuals each, which correspond to 23, 9 kg per tank.

The density in both systems was approximately  $\leq 10$  kg fish /m<sup>2</sup> and the water temperature were kept around 11 to 12 °C. The salinity in the tanks was 32-35 ‰.

### *Sea cages*

80 juvenile halibut with weights of 150 g and 60 g were set out directly into the sea cages. It was a cage within a cage, so the tested fish were not mingling with the others. The cage was hanging on a rope connected to a crane's arm, so the fish could be checked for dead ones. The water temperatures ranged from 9, 2 - 12, 5° C.

### *Procedure during the experiments*

A daily routine included; measuring the oxygen manually in the tank outlet, flushing them and collecting the dead fish.

The tanks were cleaned weakly and weight samples were taken every 2<sup>nd</sup> week. The water flow and the TGB were measured randomly during the whole experiment.

Amber Neptune (3 mm) was the chosen feed used for all tanks throughout the experiment exceptional these 20 days of feeding Antibiotics to tank 3 and 4.

Antibiotic feed (Oxilinacid) was given for 10 days at a time to 2 selected tanks, at day 2 and again at day 30 onwards during the main experiment.

Approximately 270 g feed had been distributed over each tank within 30 minutes. Over a 24 h period 900 g were fed, which corresponds to a Feed factor was set to 3. In the course of the experiment the feed amount increased according to increased biomass in the tanks.

## **3.3 Data collection**

### Water quality parameter

Water parameter were documented every week including CO<sub>2</sub>-, O<sub>2</sub>-, NH<sub>4</sub>-, NH<sub>3</sub>-Nitrate and Nitrite values.

### Chemicals

Over a period of 2 weeks dead fish were collected and their liver been removed, put together in tubes and send to the NIVA laboratory in Oslo. There the material was tested for common and chlorinated Pesticides (short: PCP, HCP).

## Growth and Mortality

To document growth and mortality weight samples are taken at the start and from then onwards every 14 days so the actual growth factor (SGR) and feed conversion ratio (FCR) can be specified.

No waste feed was collected so the feed delivered was to be assumed the feed intake.

Calculation of specific growth rate (SGR), feed conversion ratio (FCR), feed efficiency ratio (FER) and mortality (N)

**SGR (%)** =  $((\text{Endweight}/\text{Startweight})^{(1/t)} - 1) \times 100$

**FCR:** Feed intake (kg) / weight gain (kg)

**FER:** (Final weight (g) - Initial weight (g)) x Amount of Fish / Total feed eaten (g)

**Mortality (%)** =  $((N1/N0)^{(1/t)} - 1) \times 100$

V0 = initial weight

V1 = final weight period

W0 = start weight

Wn = end weight

N0 = Number of fish at start period

N1 = Number of fish after a period

BM = Biomass

SGR = Daily specific growth rate

d° = Day degrees

t = time in days

## Behaviour

The fish behaviour was observed by taking pictures and documenting abnormalities.

### **3.4 Data Analyses**

The data were analyzed with One-way ANOVA with the program Minitab provided by the Statistical institute of the UMB. The assumed model is normal distributed with  $Y_{ij} = N(\mu, \sigma)$  and it is a one way design because there is just one systematic effect in the model, the effect of treatments.

## 4. Results

### 4.1 Water parameter

Table 4.1 and 4.2 gives an overview of the water quality parameter of the A and B-recirculation systems during the experimental period. The oxygen in both systems kept above 75 % and under 105 %, which corresponds to 6, 6 mg/l and 9, 24 mg/l with a temperature of 11 to 12 °C and at a salinity of 33-35 ‰. The pH was between 7 and 7, 2 (recommendation: 7, 1 - 8, 1) and CO<sub>2</sub> between 5 to 7 mg/l, which should remain under 7 mg/l. Nitrate, Nitrite, Ammonium and Ammoniac show as well no deviation from the recommended range, though the values from the A- Hall were lower than the ones from the B - Hall and water was clearer compared to the B-Hall. This is most probably the effect of the proteinskimmer, taking out nitrogenous waste and other organic substances from the A-Hall system. Although a test for suspended solids have not been performed is it very likely that lots of particles were removed by it, visibly resulting in cleaner water.

The measurements of the gas pressure showed a slightly oversaturation of Nitrogen (N<sub>2</sub>) and were done twice during the experiment and show differences among the tanks. Oversaturation of Nitrogen is affecting the fish negative. With a mortality of 40 to 50% of the experimental fish (figure 4.2.4), it was even higher than it was usually at Bjørn. So an oversaturation could be a contributor to a higher mortality in that way that it weakens the fish.



Table 4.1: Results of the water parameter from the A- recirculation system during the experiment.

| A-Hall                 | Uke 19 | Uke 20 | Uke 21 | Uke 22 | Uke 23 | Uke 24 | Uke 25 | Uke 26 | Uke 27 | Uke 28 | Uke 29 |      |
|------------------------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|------|
| PH                     | 7,2    | 7,18   | 7,15   | 7,27   | 7,25   | 7,25   | 7,24   | 7,14   | 7,13   | 7,09   | 7,15   |      |
| NH3 NH4+               | 0,05   | 0,1    | 0,1    | 0,05   | 0,05   | 0      | 0,1    | 0,1    | 0,25   | 0,5    | 0,25   | mg/l |
| NH3                    | 0,0004 | 0,0008 | 0,0008 | 0,0004 | 0,0004 | 0,0000 | 0,0008 | 0,0008 | 0,0020 | 0,0040 | 0,0020 | mg/l |
| Nitrate for cleaning   | 10,0   | 9,0    | 10,0   | 10,0   | 6,0    | 10,0   | 8,0    | 8,0    | 9,0    | 10,0   | 8,0    | mg/l |
| Nitrate after cleaning | 44     | 39,6   | 44     | 44     | 26,4   | 44     | 35,2   | 35,2   | 39,6   | 44     | 35,2   | mg/l |
| NO2N                   | 0      | 0      | 0,05   | 0      | 0      | 0      | 0      | 0      | 0      | 0,05   | 0,05   | mg/l |
| Nitrit                 | 0      | 0      | 0,165  | 0      | 0      | 0      | 0      | 0      | 0      | 0,165  | 0,165  | mg/l |
| CO2                    | 6      | 7      | 5      | 5      | 6      | 6      | 6      | 6      | 6      | 8      | 7      | mg/l |
| Oxygen                 | 94     | 95     | 95     | 95     | 95     | 96     | 97     | 95     | 90     | 98     | 94     | %    |
| Temperatur             | 11,4   | 11,4   | 11,3   | 11,9   | 11,7   | 11,7   | 11,8   | 12     | 12,4   | 12,3   | 12,5   | °C   |
| <b>N2</b>              |        |        |        |        |        |        |        |        |        |        |        | %    |
| F3 - A+PS              | 104,9  |        |        |        |        |        |        | 105,14 |        |        |        | %    |
| F4 - A+PS              | 103,7  |        |        |        |        |        |        | 104,6  |        |        |        | %    |
| F5 - PS                | 102,13 |        |        |        |        |        |        | 103,16 |        |        |        | %    |
| F6 - PS                | 101    |        |        |        |        |        |        | 101,53 |        |        |        | %    |

NH<sub>3</sub> -Ammoniak; NH<sub>4</sub> - Amonium; CO<sub>2</sub> - Carbondioxid; NO<sub>2</sub>N - Nitrite before cleaning

A+PS - Antibiotic feed and Proteinskimmer; PS- Proteinskimmer; Control- regular feed and no Proteinskimmer.

- NO<sub>3</sub> for cleaning - samples taken before water enters the recirculation system,
- NO<sub>3</sub> after cleaning - samples taken after cleaning through recirculation system,
- NO<sub>2</sub>N -sample were taken before cleaning through recirculation system
- Recommendation: pH - 7, 1 - 8, 1; NH<sub>3</sub> - < 0, 03-0, 05 mg/l; Nitrate - < 50 mg/l; Nitrite - 0, 1-1mg/l; CO<sub>2</sub> - < 7 mg/l; N<sub>2</sub> - < 100 %.

Table 4.2: Results of the water parameter from the B- recirculation system during the experiment.

| B-Hall                 | Uke 19 | Uke 20 | Uke 21 | Uke 22 | Uke 23 | Uke 24 | Uke 25 | Uke 26 | Uke 27 | Uke 28 | Uke 29 |      |
|------------------------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|--------|------|
| PH                     | 7,01   | 7,02   | 7,02   | 7,03   | 7,04   | 7,02   | 7,02   | 7      | 7,01   | 7,15   | 7,15   |      |
| NH3 NH4+               | 0,25   | 0,25   | 0,25   | 0,5    | 0,2    | 0,2    | 0,2    | 0,1    | 0,15   | 0,5    | 0,25   | mg/l |
| NH3                    | 0,002  | 0,002  | 0,002  | 0,004  | 0,004  | 0,002  | 0,0016 | 0,0008 | 0,0012 | 0,004  | 0,002  | mg/l |
| Nitrate for cleaning   | 11     | 11     | 10     | 11     | 10     | 11     | 11     | 10     | 4      | 6      | 6      | mg/l |
| Nitrate after cleaning | 48,4   | 48,4   | 44     | 48,4   | 44     | 48,4   | 44     | 44     | 17,6   | 26,4   | 26,4   | mg/l |
| NO2N                   | 0,05   | 0,05   | 0,05   | 0,05   | 0,05   | 0,05   | 0,05   | 0      | 0,05   | 0,05   | 0,05   | mg/l |
| Nitrit                 | 0,165  | 0,165  | 0,165  | 0,165  | 0,165  | 0,165  | 0,165  | 0      | 0,165  | 0,165  | 0,165  | mg/l |
| CO2                    | 7      | 8      | 7      | 7      | 7      | 7      | 6      | 7      | 7      | 7      | 7      | mg/l |
| Oxygen                 |        | 97     | 98     | 97     | 97     | 102    | 101    | 102    | 98     | 98     | 99     | %    |
| Temperatur             | 10,7   | 10,9   | 10,8   | 10,2   | 11,7   | 11,2   | 11     | 10,8   | 11     | 11,2   | 11,1   | °C   |
| F7 - Control           | 98,6   |        |        |        |        |        |        | 101,3  |        |        |        | %    |
| F8 - Control           | 101,16 |        |        |        |        |        |        | 103,59 |        |        |        | %    |

NH<sub>3</sub> -Ammoniak; NH<sub>4</sub> - Amonium; CO<sub>2</sub> - Carbondioxid; NO<sub>2</sub>N - Nitrite before cleaning

A+PS - Antibiotic feed and Proteinskimmer; PS- Proteinskimmer; Control- regular feed and no Proteinskimmer.

- NO<sub>3</sub> for cleaning - samples taken before water enters the recirculation system,
- NO<sub>3</sub> after cleaning - samples taken after cleaning through recirculation system,
- NO<sub>2</sub>N -sample were taken before cleaning through recirculation system
- Recommendation: pH - 7, 1 - 8, 1; NH<sub>3</sub> - < 0, 03-0, 05 mg/l; Nitrate - < 50 mg/l; Nitrite - 0, 1-1mg/l; CO<sub>2</sub> - < 7 mg/l; N<sub>2</sub> - < 100 %.

#### 4.1.1 Chemicals

A chemical test was performed in order to consider the impact of possible toxic chemicals or to exclude this area entirely. For these tested chemicals are not yet limit values existing, but according to NIFES are the values in a normal range of concentration and therefore not lethal or disease causing.

Table 4.1.1 Results of the chemicals tested by NIVA in Oslo.  
Data provided by NIFES.

| Analysevariable   | µg/kg  |
|-------------------|--------|
| CB28-B            | 0.12   |
| CB52-B            | 0.11   |
| CB101-B           | <0.05  |
| CB118-B           | 0.16   |
| CB105-B           | 0.05   |
| CB153-B           | i      |
| CB138-B           | 0.26   |
| CB156-B           | 0.09   |
| CB209-B           | <0.05  |
| ΣPCB              | i<0.94 |
| ΣPCB <sub>7</sub> | i<0.79 |
| QCB-B             | 0.05   |
| HCHA-B            | <0.05  |
| HCB-B             | 0.22   |
| HCHG-B            | 0.13   |
| OCS-B             | <0.05  |
| DDEPP-B           | 0.72   |
| TDEPP-B           | 0.15   |

i- Forbindelsen er dekket av en interferens i kromatogrammet.

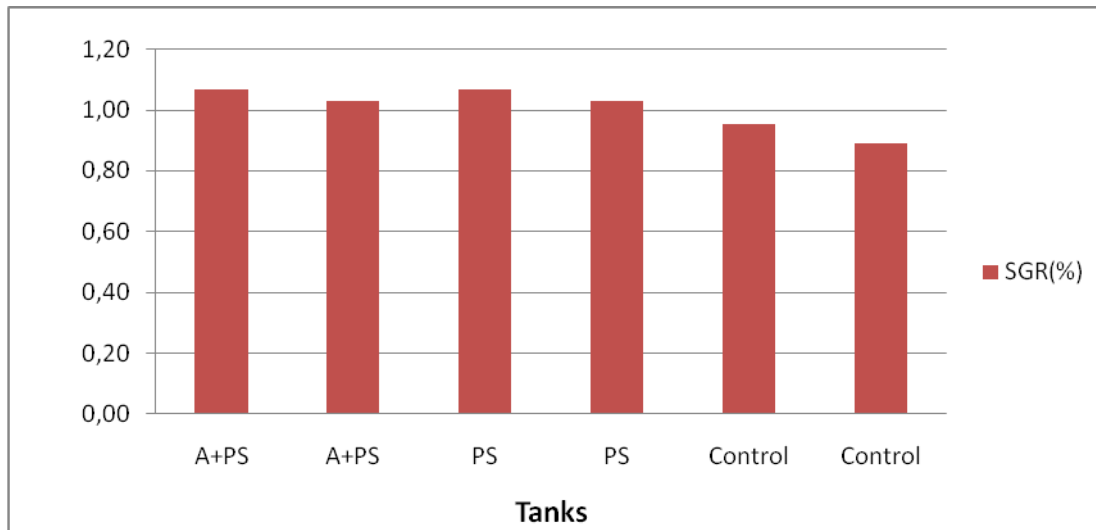
PCB - Polychlorinated biphenyls including subgroups CB...; HCB -Hexachlorbenzen; OCS - Octachlorstyren; DDEPP, TDEPP - systematic abbreviation of organochlorine insecticide

#### 4.2 Growth and Mortality

The best specific growth rate (SGR %) (Figure 4.2.1) had the fish in tanks A+PS (Antibiotic feed and Protein skimmed water), followed by tanks with PS, and the lowest growth were in tanks with regular water quality.

The first treatment with antibiotic feed and proteinskimmed water resulted in SGR values of 1,03 % and 1,07 %, second treatment with regular feed and proteinskimmed water in a SGR of 1,07 % and 1,03 % and the control group showed

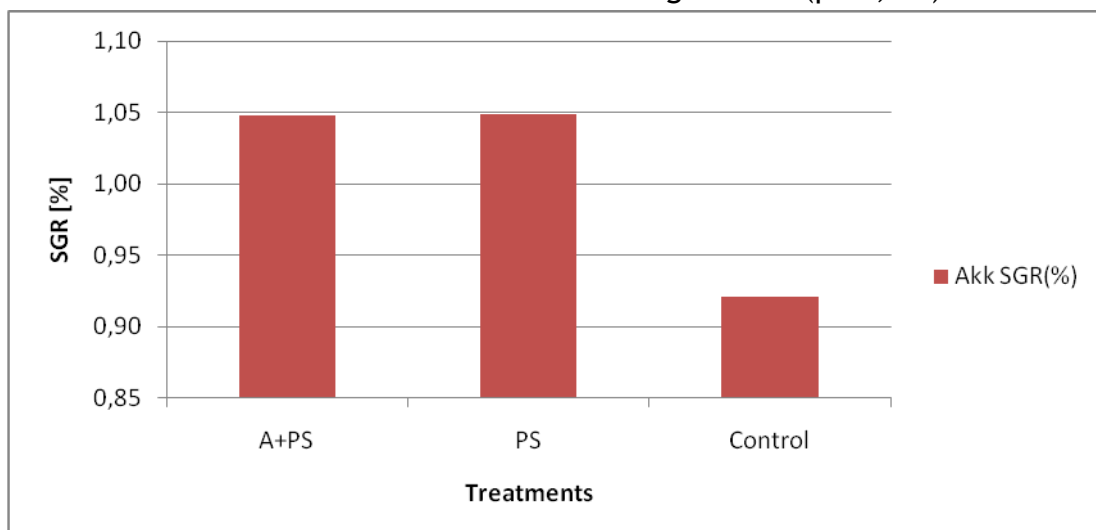
0,95 % and 0,89 % (Numbers follow the bars in the figure from left to right). Hence these differences are tank affect and show no relation towards the treatments.



A+PS - Antibiotic feed and Proteinskimmer; PS- Proteinskimmer; Control- regular feed and no Proteinskimmer.

Figure 4.2.1: Overview over the average daily specific growth rate (SGR %) for each tank during the experiment.

In average the treatment A+PS and PS had the same SGR value at the end of the experiment; which was slightly higher compared to the control group (Figure 4.2.2), showing that the Proteinskimmer had an effect on growth. However the difference between these treatments is not significant ( $p= 0, 52$ )



A+PS - Antibiotic feed and Proteinskimmer; PS- Proteinskimmer; Control- regular feed and no Proteinskimmer.

Figure 4.2.2: Average daily specific growth rate (SGR %) among the 3 different treatments.

As seen in table 4.2.1 the feed factor is around 3 in all the tanks, but the weight gain amount to 28 kg to 29 kg in the tanks with proteinskimmed water and is only

around 17 kg for the control group over a period of 119 days. The FCR in the control groups is therefore also very high which in turn explains the bad feed efficiency of 15 to 17 %. Of a good FE it is referred of values over 50 % (Steven Craig, Assistant Prof.).

A good FCR value for fish is between 1 and 2, but data here show values from 4 to even 6, while the weight gain is remains under average.

Table 4.2.1: Overview over total feed intake, feed factor, feed conversion ratio, feed efficiency and weight gain over the whole experimental period.

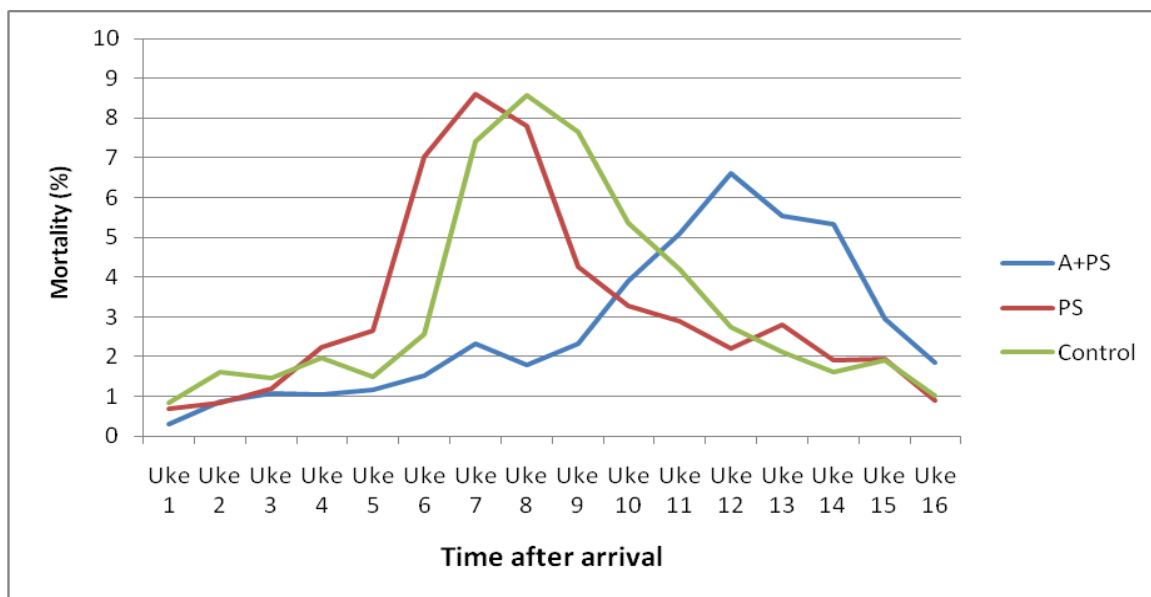
| Treatments   | Feed delivered (Kg) | Feed Factor | FCR  | FE (%) | Weight gain (kg) |
|--------------|---------------------|-------------|------|--------|------------------|
| A+PS (F3)    | 131                 | 3,0         | 4,49 | 22,29  | 29,20            |
| A+PS (F4)    | 141,7               | 3,0         | 4,93 | 20,30  | 28,76            |
| PS (F6)      | 128,2               | 3,0         | 4,53 | 22,08  | 28,30            |
| PS (F5)      | 123,5               | 3,0         | 5,69 | 17,58  | 21,71            |
| Control (F7) | 103,1               | 3,0         | 5,75 | 17,38  | 17,92            |
| Control (F7) | 110,3               | 3,0         | 6,39 | 15,64  | 17,25            |

A+PS - Antibiotic feed and Proteinskimmer; PS- Proteinskimmer; Control- regular feed and no Proteinskimmer.

FCR - Feed conversion ratio, FE (%) - Feed efficiency

Both the fish with Proteinskimmer and without showed a raise in mortality after week 5 or 6 and returned to the previous number of dead fish at week 12 / 13, although the increase is steeper than the drop.

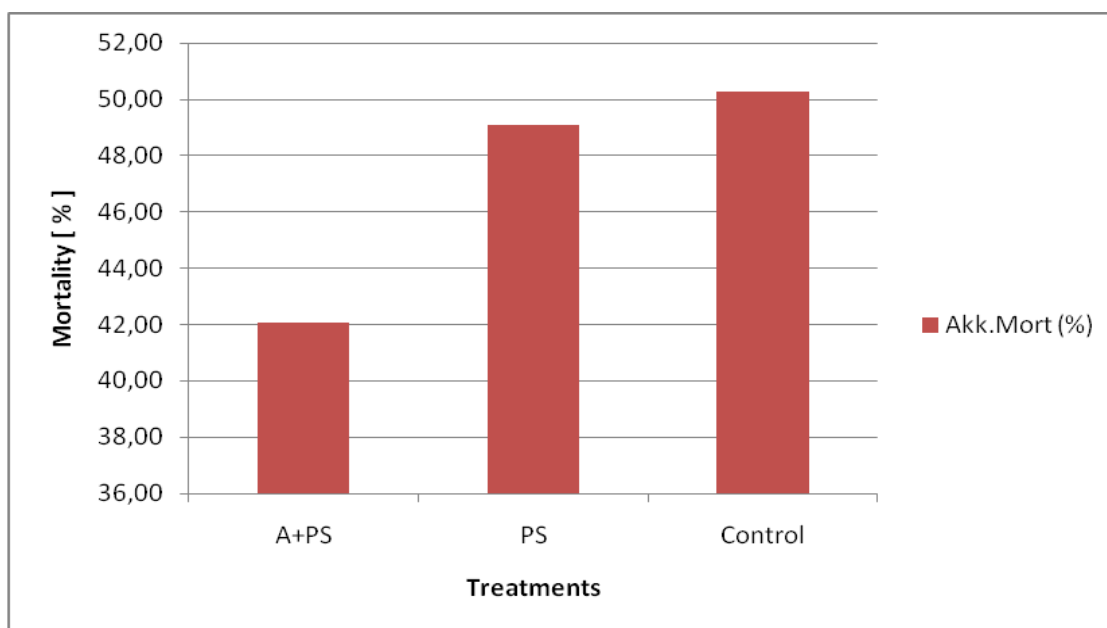
While treatment PS and the control group showed a raise in mortality at week 5 was it with treatment A+PS around week 9 after arrival. Not only is the curve shifted but also less high and the increase as well as the drop is gradually (Figure 4.2.3).



A+PS - Antibiotic feed and Proteinskimmer; PS- Proteinskimmer; Control- regular feed and no Proteinskimmer.

Figure 4.2.3: Mortality development of all 3 different treatments.

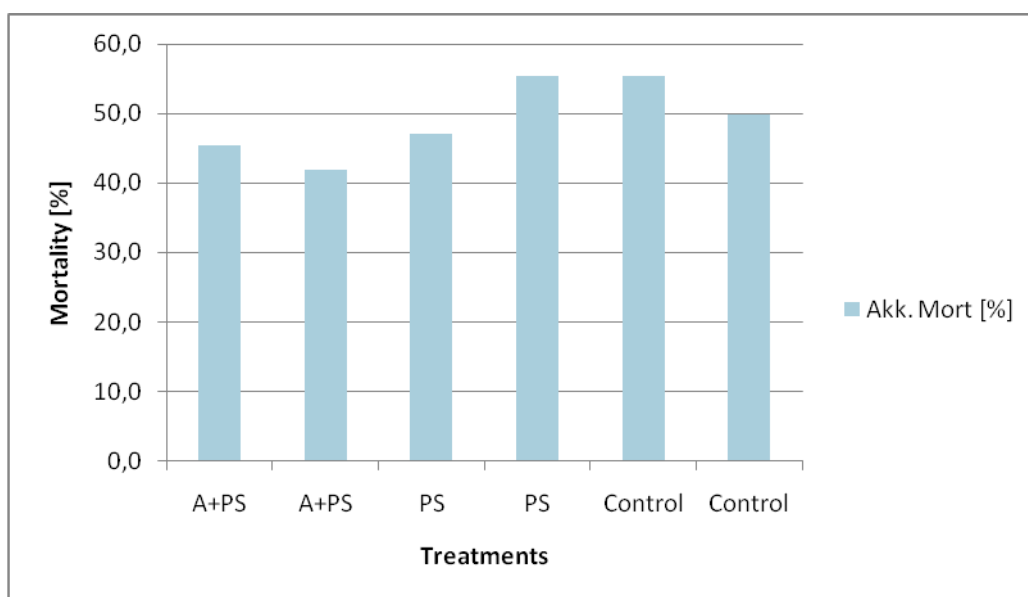
But when looking at the accumulated mortalities (Figure 4.2.4) between the 3 different treatments it shows that there is no significance between proteinskimmed (PS)- and regular recirculated marine water (Control) and also the difference between treatment (PS) and treatment (A+PS) is not significant ( $p= 0,76$ ), therefore had the Proteinskimmer no effect on the mortality extent.



A+PS - Antibiotic feed and Proteinskimmer; PS- Proteinskimmer; Control- regular feed and no Proteinskimmer.

Figure 4.2.4: Accumulated Mortality after completing the experiment.

When looking at single accumulated mortalities of each tank, is it with Number 1 43 % and Number 3 with 45 %, which shows that Antibiotic feed did not reduce the amount of dead fish compared to regular fed fish. The same is the case with tank 4 (52, 9 %) and 5 (52, 8 %), even better water quality had higher mortalities than the control groups. Confirming the statements from above that the treatments had no effect and these differences (Figure 4.2.5) are more or less due to tank effects. (Numbers are to be counted from left to right)



A+PS - Antibiotic feed and Proteinskimmer; PS- Proteinskimmer; Control- regular feed and no Proteinskimmer.

Figure 4.2.5: Mortality variations among the tanks.

### Sea Cages

The fish, which were set out into the sea cages had a high mortality and a slow growth rate (figure 4.2.2), which was mainly due to improper feeding and looking after. Therefore the data are not useful for a comparison with the landbased experiment. Since the fish died and did not grow because of lack of feed. So the feed factor is so dominant that the influence of other factors towards mortality and growth cannot be assumed.

Table 4.2.2: Results from the fish held in the sea cages during the experiment.

| Set in /check up | 28. April | 28. April | 22.Juni | 22.Juni |
|------------------|-----------|-----------|---------|---------|
| Amount           | 150       | 150       | 101     | 103     |
| Weight [g]       | 16        | 43        | 16,8    | 47,9    |
| Mortality. [%]   |           |           | 32,7    | 31,3    |
| SGR [%]          |           |           | 0,09    | 0,2     |

### 4.3 Behaviour

For Halibut it is considered a normal behaviour to lay down on the bottom, most fish did that after setting them into the tanks. Only after approximately 2 weeks some fish started to swim at the surface and started nipping, which looks like as if the fish snaps for air. When offering feed these fish don't eat even when feed is offered right in front of them.

#### *Eating*

In the experiment it was observed that when halibut feed they focus the feed pellet first and then quickly "attack" which is not always successful, sometimes they miss the feed. So the fish have to start all over again; approaching - focusing and attacking. But Halibut do both eat swimming pellets but also the ones lying on the bottom.

#### *Aggressiveness*

When feed was offered halibut seem to be getting aggressive, some for example try to mark their territory by waving their tale and swimming in a circle and chasing others away. Others turn up-side-down and try to lift up other fish from their laying spot. The repressed fish, when not finding other spots to lie down begin to swim. Biting one another is common among halibut also, especially bigger ones attack smaller also when there is no feeding time. Consequently quite an amount of fish had injuries especially the eyes are affected, but also the fins. Injured fish are stressed and show reduced appetite.

## 5. Discussion

The high mortalities at Bjørn existed since the beginning of the ongrowing site of Aga Halibut. All attempts to solve the problem gave no specific results and no indication in what direction to investigate further. The set up of the experiment was therefore much aligned and it was mainly the goal to narrowing down the problem. A comparison of different treatments was after all considerations the best way to do so and to make statements on water quality and possible Bacteria or other microorganism infestation.

The experiment overall went as planed. The pre- experiment was helpful in that sense that technical-, set up- and performance problems were discovered and corrected. The pre- experiment ended after 4 weeks due to new arriving experimental fish from Risør. The results of it, however, were not used because the fish were put together in one of the production tanks and were later on mingled with other fish, therefore it was not possible to track the different treated fish groups. The only problem during the main experiment was the sea cage part. Due to organisation problems the fish received not enough feed, probably causing the death of many individuals as well as the non-growth. Additionally was it started 17 days later and was ended 14 days earlier, which in turn led to variation and thereby to a poor comparison basis. But it had no effect on the experiment itself. Neither the Proteinskimmer nor the use of Antibiotics had effects on growth and/or mortality. Though the results show a small improvement, are they not significant leading to the idea that the water quality, regarding physical and organic components, is not the source of the problem. A certain bacteria or virus infection could be considered but is difficult to underpin; all bacteria known to be causing disease in Halibut have been checked, also have they been found randomly but as mentioned earlier the amount was too low in order to cause diseases or mortalities, as in general a bacteria problem occurs with poor hygiene, which was not the case at Bjørn. Two viruses, IPN and Nodavirus, have been found negative. Though others have not been looked for, veterinarian examination didn't find symptoms or any other hints to support this idea. Unless a totally unknown virus or other micro-organisms have been developed being difficult to identify. However, certain symptoms regarding inflammation, organ modification or any other failures would have been noted.

To pinpoint the mortality issue 2 possibilities can be considered; stress and heavy metals.

Stress leads to reduced immune response and/or reduced feed uptake. A characteristic feature of the behavioral response to intensely acute or chronic stressors is a reduction in appetite. In fish, as in other vertebrates, the corticotrophin-releasing factor (CRF) system plays a key role in coordinating the ... behavioral responses to stress.... In addition, the appetite suppressing effects of



various environmental, pathological, physical, and social stressors are associated with elevated levels of forebrain CRF ... and with an activation of the hypothalamic-pituitary-interrenal (HPI) stress axis [Bernier 2006]. The long transport in connection with changing temperatures could be one reason for developing such severe stress, but moreover is the social interaction of the halibut itself a central problem. Unlike salmon, for example, are halibuts territorial, so they prefer to lay down on the bottom acquiring space for themselves or in groups. As described earlier are especially small ones known to be most aggressive [Greaves and Tuene 2000], compared to later stages in life. In addition to that the growth among the fish is very uneven, bigger fish will compete for more space, forcing smaller fish to swim, which consequently have less opportunities to feed. Ones the feed uptake is reduced, fish become smaller and again feed uptake becomes more difficult, since attacking requires concentration and thereby energy. In the same way are injured fish stressed and consequently show reduced appetite but additionally have difficulties in focusing the feed pellet i.e. when eyes are injured or missing. But neither the social aggressiveness nor the transports can be totally responsible for the high mortalities at Bjørn, especially so similar from fish group to fish group. There is a strong indication that the fish suffered from a metal poisoning, more precisely copper toxification.

First all fish no matter what size that came to Bjørn, showed the same abnormal behaviour; gasping for air at the water surface, swimming continuously in the tanks, being apatic and reduced feed uptake.

Secondly the diagnosis made by the veterinarian, liver and gill hypertrophy and even gill inflammation are strongly indicating that these organs were hyperactive and overworked. With an oversupply the body will still uptake copper, since it is an important element for many functions in the organism. As described by Grosell and Wood 2002, fish accumulate higher levels of copper in the liver and gills, disturbing metabolic functions and possibly leading to a hypertrophy of these organs.

According to McGeer et al. 1999, high copper levels reduce the appétit, which was observed regularly at Bjørn and which is understandable with a metabolic dysfunction. Altogether it gives an explanation to the overall low growth rate, even after installing the proteinskimmer. Furthermore a higher copper level in gills or liver is difficult to detect, even during veterinarian examination it can be overseen, when it is not specifically looked for.

Thirdly the shifted peak of the mortality curve of the fish, treated with Antibiotic feed. A higher copper accumulation will lead to an imbalance between copper and zinc reducing the immune response and making the fish prone for any kind of infections [Dr. Wilson, 2011]. The antibiotic in this context delayed the outbreak of any infection.

And perhaps the strongest indication is the critical amount of copper (2, 2 µg/l) detected in the recirculation water in 2007. The metals were not detected high in

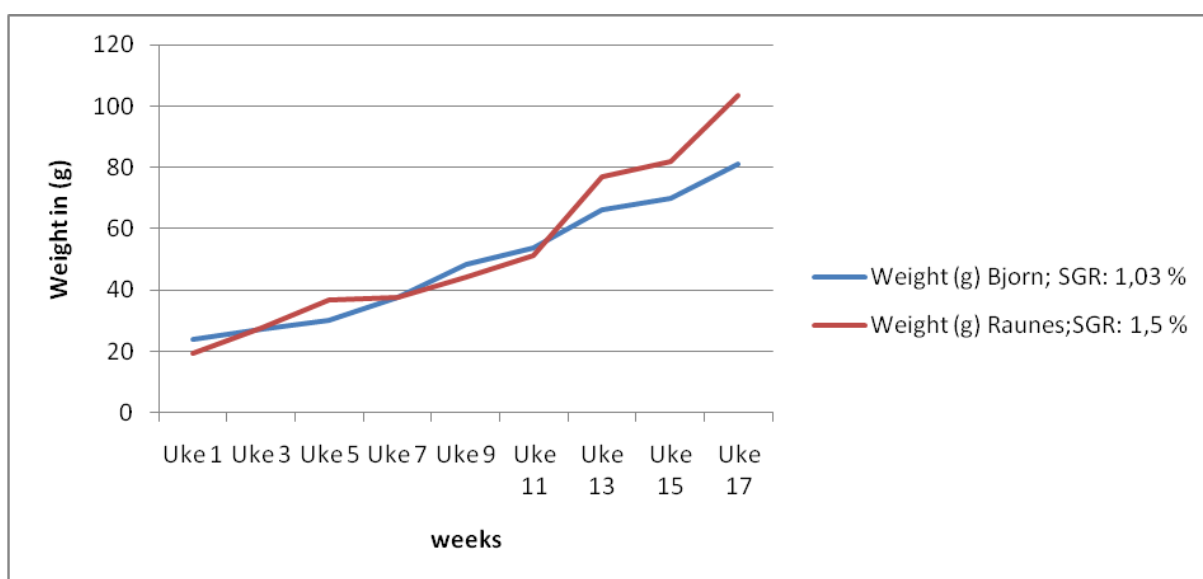
the intake but recirculation water, meaning that the source of it lies within the facility, which in turn led to accumulation of them over time especially with a high recirculation factor of 98%. Although the propeller of the main pumps had been coated with a ceramic layer, is there always a possibility of metal oxidation. Copper as other base metals react quickly in alkali solution (saltwater) forming together with oxygen copper (I) and copper (II) oxides, diluting copper ions into the water. Higher temperatures and alkalinity enhance this process. In addition a ceramic layer applied over the propellers is not a guarantee of absolute protection against metal leakage [www.patentstorm.us]. The composition as well as the way of application of the ceramic layer is decisive for the reliability of the layer. Since ceramic is not a totally dense material and since it was exposed to mechanical forces, occurrence of porosity, corrosion and breakages has to be considered. One factor, speaking against a copper problem is the fact that in a flow- through system the metals cannot accumulate. With a retention time of 40 minutes, it is very unlikely that copper will be stored in the water body affecting fish. But 3 features of copper should be seen; copper in low amounts are already toxic to aquatic organism [Riedel 2008], copper as an essential trace element will be taken up by the organism and hence will accumulate [McGeer et al. 1999] plus little is known about the tolerance range of higher copper levels in halibut and thirdly, copper as other heavy metals have a seasonal occurrence. The test for heavy metals, made 2007 is actually not very representative. It is always advisable to test over a longer period of time, since also the metal occurrence in the sea have seasonal fluctuations[Conan and Stengel, 2011].The same applies for the metals released within the plant. Since it is dependent on water properties like temperature, organic substances, etc. freeing of metal ions is fluctuating as well. In the flow- through system the mortality occurrence was low, around 5% and high, around 24%, confirming the idea that higher amount of free copper ions released from the pumps could be at times more and other times less, which makes it so important to check for metals more often, in order to obtain reliable results. In turn, these facts enhance also the possibility of metals coming from the sea. And since the symptoms of the fish in the flow -through- and recirculating system is so similar it must be assumed that the problem has the same origin in both system, being either the pump`s propeller and/or metals coming from the environment. However, it should be mentioned that the finding of a hidden metal problem was discovered after the experiment was already over and since the running costs of the farm were exceeding their income, it was decided to close down the facility and all fish were shifted out to the sea cages.

After close down, the new juveniles from Risør were moved to a totally different place, called Raunes, which is a simple flow- through system. It was of interest if the problem was farm or fish related.

A comparison was done for the growth, the FCR (feed conversion ratio) and the mortality. To make the comparison suitable fish data with same weights had been selected:

### Growth

Figure 5.1 demonstrates the weight gain of the same fish size from set in until after 2 months. Fish from Bjørn need obviously more time to growth than at e.g. Raunes.



Source: Bjørn: Example growth of one of the fish tanks of the experiment.

Raunes: Example growth of one of the fish tanks at Raunes Fiske Farm.

Figure 5.1: Comparison of two growing curves of small on- growing Halibut at two different facilities over a period of 17 weeks. Flow - through system (Raunes) and recirculation system (Bjørn).

### FCR

Secondly the FCR values from Bjørn are much higher compared to Raunes. Figure 5.2 gives an idea about reasonable values of 1, 3 to 1, 7 at Raunes and abnormally high for Bjørn (4, 9 to 6, 3), at least for fish. This implies that halibut would need around 5 kg feed in order to put on 1 kg of weight, but these values are close to a FCR of ruminants, which are around 7, however that cannot be the case, it rather confirms that the fish show a low appétit and lots of feed had been uneaten, resulting in a poor growth.

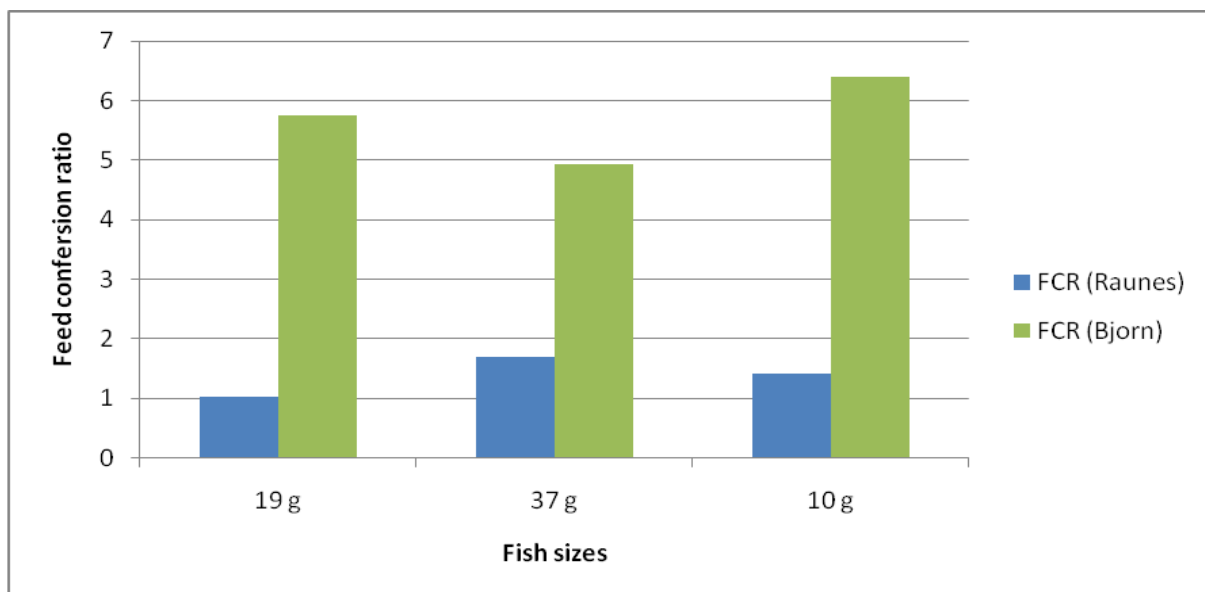


Figure 5.2: Comparison of the FCR values of the same fish sizes at two different facilities; with flow - through system Raunes and recirculation system (Bjørn).

### Mortality

Figure 5.3 shows the differences in mortality after a period of 16 to 17 weeks. It is more than abnormal with mortalities of 40 % and higher, when it is around 2 % to 4 % at another site (e.g. Raunes) with the same type of fish.

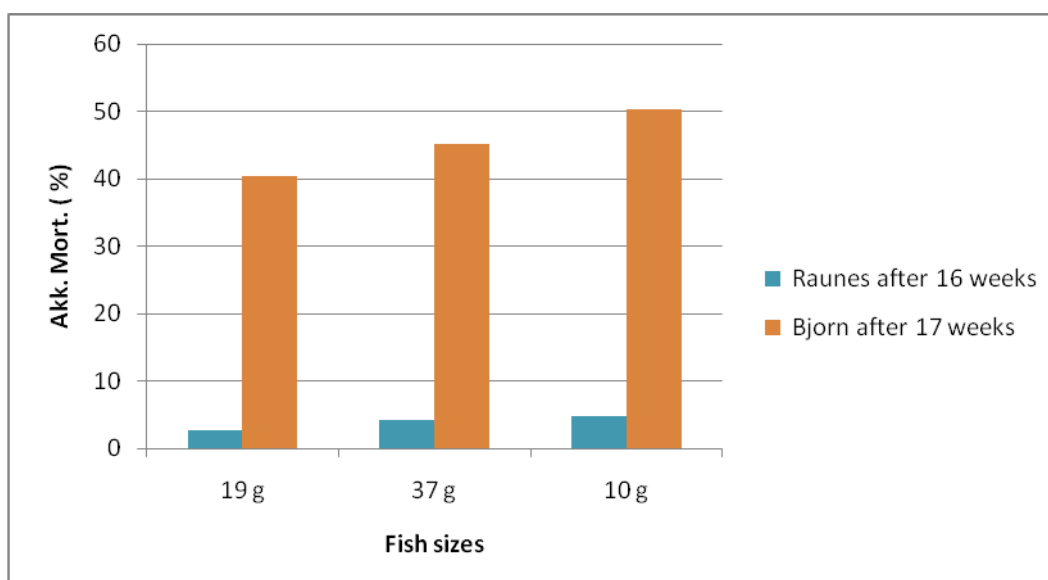


Figure 5.3: Comparison of summarized mortalities of two different facilities raising juvenile Atlantic Halibut; with flow - through system Raunes and recirculation system (Bjørn).

These comparisons are not necessarily indicating a copper or metal toxication but they underline, that the fish at Bjørn were not in a healthy environment. Observing

and working with the fish at Raunes together with all the findings raised the idea of a possible metal problem.

In a future experiment it would be focused on discovering any kind of metals hidden within the system. This can be done without using fish performances as an indicator but taking water samples which are tested for heavy metals before and after possible water metal - contact devices, including pumps, filters etc. In order to be positive effected fish have to be tested for heavy metal accumulation and tolerance by examining the liver and gills, but also metal balances in general, which would be very informative towards application of equipment.

## 6. Conclusion

In conclusion the question if hidden bacteria or other micro-organism as a possible reason for the high mortalities can be negated. And if a better water quality can improve the growth rate and/or lower the mortality can be actually affirmed. The reason for the high mortalities and the low growth rate must be seen in connection and searched within the water quality. Although the results of the experiment show no effect in any of these treatments and are therefore not directly useful, confirm they the findings of earlier documentation and reports, which point towards a metal intoxicification, which as a possible reason, has been totally neglected.

The amount of copper found in the water may not be acutely dangerous but definitely weakening the fish to such an extent that secondary factor such as stress, and injuries cause death.

The low growth rate confirms this thesis more than considering that a recirculation system for fish farming is not suitable. A total check of the plant for metal-water contact would therefore be recommended, also with the fact that the previous production of cod failed as well.

The purpose of the experiment was thereby fulfilled, because the problem was not just narrowed down but leading to a strong indication of heavy metal pollution.

### **Acknowledgment**

Herby would I like to thank all who contributed to the development of this work. First of all Odd- Ivar Lekang, as supervising project responsible and Bjørn Frode Erikson, as co-supervisor for correction of the work, guidance through the experimental period and advices for improvements. Further I would like to mention Aga Halibut for provision of the project and employees, especially Stig Hjordhal, Trude Hagen, Jonny Innvær and Yuri Marchenko for construction of the experimental system, assistance of collecting data and support with any kinds of problems.

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*Can Proteinskimmers improve water quality in recirculation systems so that it effects Growth and Mortality of juvenile Atlantic Halibut (Hippoglossus hippoglossus) and can Antibiotic feed help to uncover hidden Bacteria?*

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**2011**

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**Appendix**

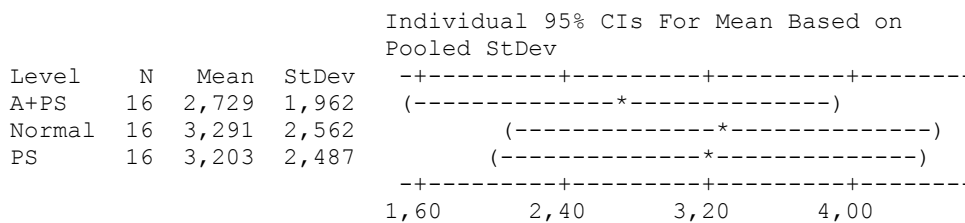
Statistic

The output from the Mortality is as followed:

**Mortality (%) versus Treatment**

| Source    | DF | SS     | MS   | F    | P     |
|-----------|----|--------|------|------|-------|
| Treatment | 2  | 2,92   | 1,46 | 0,26 | 0,769 |
| Error     | 45 | 248,91 | 5,53 |      |       |
| Total     | 47 | 251,83 |      |      |       |

S = 2,352    R-Sq = 1,16%    R-Sq(adj) = 0,00%



Grouping Information Using Fisher Method

| Treatment | N  | Mean  | Grouping |
|-----------|----|-------|----------|
| Normal    | 16 | 3,291 | A        |
| PS        | 16 | 3,203 | A        |
| A+PS      | 16 | 2,729 | A        |

All means share the same letter, showing that there is no significantly difference among the treatments. The test was done at a 95 % confidence interval, and because  $p = 0,76 > \alpha = 0,05$  is Hypothesis 1 rejected at a 5 % level of significance.

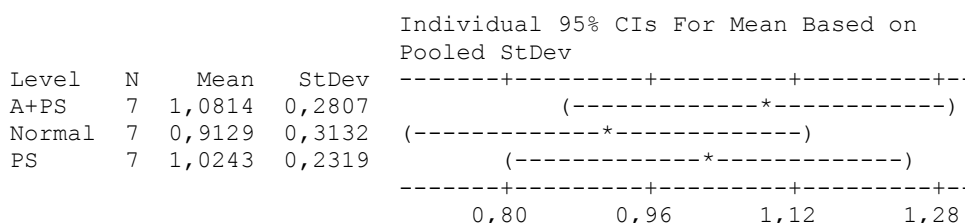
**The Hypothesis:**

Nullhypothesis =  $H_0: \mu_1 = \mu_2 = \mu_3$  against  $H_1: \text{At least one } \mu_i \text{ is different from the other population means.}$

**SGR (%) versus treatment**

| Source   | DF | SS     | MS     | F    | P     |
|----------|----|--------|--------|------|-------|
| Treat._1 | 2  | 0,1029 | 0,0514 | 0,67 | 0,524 |
| Error    | 18 | 1,3840 | 0,0769 |      |       |
| Total    | 20 | 1,4869 |        |      |       |

S = 0,2773    R-Sq = 6,92%    R-Sq(adj) = 0,00%



Pooled StDev = 0,2773

Grouping Information Using Fisher Method

| Treat._1 | N | Mean   | Grouping |
|----------|---|--------|----------|
| A+PS     | 7 | 1,0814 | A        |
| PS       | 7 | 1,0243 | A        |
| Normal   | 7 | 0,9129 | A        |

All means share the same letter, showing that there is no significantly difference among the treatments. The test was done at a 95 % confidence interval, and because  $p = 0,52 > \alpha = 0,05$  is Hypothesis 1 rejected at a 5 % level of significance

Table of veterinarien diagnosis made during the years 2004 until 2010, by Bjørn - Inge Rickardsen, Fiskehelse tjenneste, Dønna.

| Dato     | Slags undersokri | Skader            | Felipigmentering | Bakterier  | Parasiter             | Hypertrophie Liver | Hypertrophie Gjeller |
|----------|------------------|-------------------|------------------|--|-----------------------|--------------------|----------------------|
| 2004/1   | alle             | oye,finnsilitasje | x                | Trichodina,beveglige bakterier                     |                       |                    |                      |
| 2004/3   | dode, svimer     | oye,finnsilitasje | x                | Trichodina,beveglige bakterier                     |                       |                    |                      |
| 2004/4   | dode, svimer     | oye,finnsilitasje | x                | ingen  |                       |                    |                      |
| 2004/5   | tilfeldig utvalg | oye,finnsilitasje | x                | Trichodina,beveglige bakterier                     |                       |                    |                      |
| 2005/10  | tilfeldig utvalg | oye,finnsilitasje | x                | Trichodina,beveglige bakterier                     |                       |                    | x                    |
| 2005/11  | tilfeldig utvalg | oye,finnsilitasje | x                | ingen  |                       |                    |                      |
| 2006/01  | dode             |                   | x                | ingen  |                       | x                  |                      |
| 2006/02  | tilfeldig utvalg | oye,finnsilitasje | x                | Trichodina A-Hall                                  |                       |                    |                      |
| 2007/1   | dode             |                   | x                | Trichodina, atypisk Aeromonas                      |                       | x                  | x                    |
| 2007/2   | dode, svimer     | oye,finnsilitasje | x                | ingen  |                       | x                  |                      |
| 2007/9   | dode, svimer     | oye,finnsilitasje | x                | infecction med Ichthyobodo necator gjeller(Costia) |                       |                    |                      |
| 2008/4   | dode, svimer     | ikke dokumentert  | ikke dokumentert | atypisk furunkulose                                | entodella hippoglos;x | x                  |                      |
| 2009/4   | dode, svimer     | ikke dokumentert  | ikke dokumentert | Costia   |                       | x                  |                      |
| 2009/5   | dode, svimer     | ikke dokumentert  | ikke dokumentert | Costia   |                       | x                  |                      |
| 2009/11+ | dode, svimer     | ikke dokumentert  | ikke dokumentert | ingen  |                       | x                  |                      |
| 2010/1   | dode, svimer     | ikke dokumentert  | ikke dokumentert | korte, kokkoide, ingen Costia                      |                       | x                  |                      |
| 2010/1   | dode, svimer     | ikke dokumentert  | ikke dokumentert | bakteriehopet                                      |                       | x                  |                      |
| 2010/2   | dode, svimer     | ikke dokumentert  | ikke dokumentert | kort, stavformede Bakteria, atypisk                | parasitaer gjellbeter | x                  |                      |
| 2010/5   |                  | ikke dokumentert  | ikke dokumentert | atypisk furunkulose                                |                       |                    |                      |

Data collected form Pre-experiment from Bjorn:

| Uke 13 | Amount | Mortality[%] | Weight [g] | Biomass[kg] | O2[%]                      | Temp[C]   |             |         |
|--------|--------|--------------|------------|-------------|----------------------------|-----------|-------------|---------|
| F3     | 1275   |              | 25,11      | 32          | 96,85,94                   | 11        |             |         |
| F4     | 1057   | 1            | 30,29      | 32          | 100,88,78                  | 11        |             |         |
| F5     | 1037   |              | 30,87      | 32          | 100,96,94                  | 11        |             |         |
| F6     | 1308   |              | 24,46      | 32          | 101,93,68                  | 11        |             |         |
| F7     | 1001   |              | 31,96      | 32          | 95,76,90                   | 11        |             |         |
| F8     | 1041   | 1            | 30,75      | 32          | 93,77,94                   | 11        |             |         |
|        |        |              |            |             |                            |           |             |         |
| Uke 14 | Amount | Mortality    | Weight [g] | Biomass[kg] | O2[%]                      | Temp[C]   |             |         |
| F3     | 1275   | 2            | 27,11      | 34,6        | 102,100,100,101,101,102    | 11,3-11,6 |             |         |
| F4     | 1056   | 3            | 29,78      | 31,4        | 105,101,101,102,99,102,102 | 11,3-11,6 |             |         |
| F5     | 1037   | 2            | 32,42      | 33,6        | 105,104,104,103,102,99,101 | 11,3-11,6 |             |         |
| F6     | 1308   | 4            | 26,4       | 34,5        | 105,101,97,96,96,97,96     | 11,3-11,6 |             |         |
| F7     | 1001   | 1            | 32,81      | 32,8        | 76,95,92,96,94,94,92       | 11,1-11,3 |             |         |
| F8     | 1040   | 3            | 35,34      | 36,7        | 75,95,94,98,93,93,92       | 11,1-11,3 |             |         |
| Uke 15 | Amount | Mortality    | Weight [g] | Biomass[kg] | O2[%]                      | Temp[C]   |             |         |
| F3     | 1273   | 0            | 27,11      | 34,5        | 98-101                     | 11,6      |             |         |
| F4     | 1053   | 2            | 29,78      | 31,4        | 102-105                    | 11,6      |             |         |
| F5     | 1035   | 1            | 32,42      | 33,6        | 96-100                     | 11,6      |             |         |
| F6     | 1304   | 3            | 26,4       | 34,4        | 96                         | 11,6      |             |         |
| F7     | 1000   | 0            | 32,81      | 32,8        | 86-91                      | 11,3      |             |         |
| F8     | 1037   | 3            | 35,34      | 36,6        | 86-93                      | 11,3      |             |         |
| Uke 16 | Amount | Mortality    | Weight [g] | Biomass[kg] | O2[%]                      | Temp[C]   | SGR(%)      | Mort(%) |
| F3     | 1273   | 2            | 32,44      | 41,30       | 96-102                     | 11,4(7,4) | <b>1,20</b> | 0,16    |
| F4     | 1051   | 4            | 33,25      | 34,95       | 100-103                    | 11,4(7,4) | <b>0,73</b> | 0,38    |
| F5     | 1034   | 5            | 42,1       | 43,53       | 96-100                     | 11,4(7,4) | <b>1,74</b> | 0,48    |
| F6     | 1301   | 3            | 32,79      | 42,66       | 95-97                      | 11,4(7,4) | <b>1,45</b> | 0,23    |
| F7     | 1000   | 3            | 37,76      | 37,76       | 89-93                      | 11,2      | <b>0,94</b> | 0,30    |
| F8     | 1034   | 7            | 36,65      | 37,88       | 89-92                      | 11,2      | <b>0,24</b> | 0,68    |

Data: Mortality and SGR collected at Bjorn

| Uke 17 | Amount | Mortality | Weight [g] | Biomass[kg] | O2[%] | Temp[C] | Mort(%) | SGR(%) |
|--------|--------|-----------|------------|-------------|-------|---------|---------|--------|
| F3     | 1069   | 0         | 22,35      | 23,9        | 108   | 11,3    |         |        |
| F4     | 997    | 0         | 23,96      | 23,9        | 108   | 11,3    |         |        |
| F5     | 1018   | 1         | 23,46      | 23,9        | 107   | 11,3    |         |        |
| F6     | 1009   | 1         | 23,65      | 23,9        | 106   | 11,3    |         |        |
| F7     | 1231   | 0         | 19,4       | 23,9        | 101   | 10,9    |         |        |
| F8     | 1269   | 0         | 18,83      | 23,9        | 99    | 10,9    |         |        |
|        |        |           |            |             |       |         |         |        |



| Uke 18 | Amount | Mortality | Weight [g] | Biomass[kg] | O2 [%] | Temp[C] | Mort(%) | SGR(%)  |
|--------|--------|-----------|------------|-------------|--------|---------|---------|---------|
| F3     | 1069   | 3         | 22,35      | 23,9        | 102    | 11,1    | 0,28    |         |
| F4     | 997    | 3         | 23,96      | 23,9        | 102    | 11,1    | 0,30    |         |
| F5     | 1017   | 5         | 23,46      | 23,9        | 101    | 11,1    | 0,49    |         |
| F6     | 1008   | 9         | 23,65      | 23,8        | 101    | 11,1    | 0,89    |         |
| F7     | 1231   | 11        | 19,4       | 23,9        | 100    | 10,4    | 0,89    |         |
| F8     | 1269   | 10        | 18,83      | 23,9        | 100    | 10,4    | 0,79    |         |
|        |        |           |            |             |        |         |         |         |
| Uke 19 | Amount | Mortality | Weight [g] | Biomass[kg] | O2 [%] | Temp[C] | Mort(%) | SGR(%)  |
| F3     | 1066   | 13        | 27,03      | 28,81       | 94     | 11,4    | 1,22    | 1,19    |
| F4     | 994    | 5         | 27,22      | 27,06       | 94     | 11,4    | 0,50    | 0,80    |
| F5     | 1012   | 7         | 26,81      | 27,13       | 94     | 11,4    | 0,69    | 0,83    |
| F6     | 999    | 10        | 25         | 24,98       | 93     | 11,4    | 1,00    | 0,35    |
| F7     | 1220   | 19        | 20,46      | 24,96       | 99     | 10,7    | 1,56    | 0,33    |
| F8     | 1259   | 21        | 21,6       | 27,19       | 99     | 10,7    | 1,67    | 0,86    |
|        |        |           |            |             |        |         |         |         |
| Uke 20 | Amount | Mortality | Weight [g] | Biomass[kg] | O2[%]  | Temp[C] | Mort(%) | SGR(%)  |
| F3     | 1053   | 10        | 27,03      | 28,5        | 95     | 11,4    | 0,95    |         |
| F4     | 989    | 12        | 27,22      | 26,9        | 95     | 11,4    | 1,21    |         |
| F5     | 1005   | 13        | 26,81      | 26,9        | 94     | 11,4    | 1,29    |         |
| F6     | 989    | 11        | 25         | 24,7        | 96     | 11,4    | 1,11    |         |
| F7     | 1201   | 19        | 20,46      | 24,6        | 99     | 10,8    | 1,58    |         |
| F8     | 1238   | 17        | 21,6       | 26,7        | 99     | 10,8    | 1,37    |         |
|        |        |           |            |             |        |         |         |         |
| Uke 21 | Amount | Mortality | Weight [g] | Biomass[kg] | O2[%]  | Temp[C] | SGR(%)  | Mort(%) |
| F3     | 1043   | 12        | 29,27      | 30,5        | 96     | 11,4    | 0,61    | 1,15    |
| F4     | 977    | 9         | 30,1       | 29,4        | 94     | 11,4    | 0,77    | 0,92    |
| F5     | 992    | 25        | 28,81      | 28,6        | 95     | 11,4    | 0,55    | 2,52    |
| F6     | 978    | 19        | 30,32      | 29,7        | 95     | 11,4    | 1,48    | 1,94    |
| F7     | 1182   | 25        | 22,87      | 27,0        | 97     | 10,8    | 0,86    | 2,12    |
| F8     | 1221   | 22        | 23,82      | 29,1        | 93     | 10,8    | 0,75    | 1,80    |
|        |        |           |            |             |        |         |         |         |
| Uke 22 | Amount | Mortality | Weight [g] | Biomass[kg] | O2[%]  | Temp[C] | SGR(%)  | Mort(%) |
| F3     | 1031   | 10        | 29,27      | 30,2        | 95     | 11,3    |         | 0,97    |
| F4     | 968    | 13        | 30,1       | 29,1        | 95     | 11,3    |         | 1,34    |
| F5     | 967    | 25        | 28,81      | 27,9        | 95     | 11,3    |         | 2,59    |
| F6     | 959    | 26        | 30,32      | 29,1        | 95     | 11,3    |         | 2,71    |
| F7     | 1157   | 18        | 22,87      | 26,5        | 98     | 10,4    |         | 1,56    |
| F8     | 1199   | 17        | 23,82      | 28,6        | 97     | 10,4    |         | 1,42    |

| <b>Uke 23</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
|---------------|---------------|------------------|-------------------|--------------------|--------------|----------------|---------------|----------------|
| F3            | 1021          | 18               | 35,04             | 35,78              | 91           | 11,9           | 1,29          | 1,76           |
| F4            | 955           | 12               | 37,5              | 35,81              | 91           | 11,9           | 1,57          | 1,26           |
| F5            | 942           | 73               | 34,12             | 32,14              | 94           | 11,9           | 1,21          | 7,75           |
| F6            | 933           | 59               | 33,74             | 31,48              | 94           | 11,9           | 0,76          | 6,32           |
| F7            | 1139          | 35               | 24,87             | 28,33              | 96,5         | 11             | 0,60          | 3,07           |
| F8            | 1182          | 24               | 25,83             | 30,53              | 96           | 11             | 0,58          | 2,03           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 24</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 1003          | 22               | 35,04             | 35,1               | 89           | 11,7           |               | 2,19           |
| F4            | 943           | 23               | 37,5              | 35,4               | 94           | 11,7           |               | 2,44           |
| F5            | 869           | 61               | 34,12             | 29,7               | 97           | 11,7           |               | 7,02           |
| F6            | 874           | 89               | 33,74             | 29,5               | 97           | 11,7           |               | 10,18          |
| F7            | 1104          | 92               | 24,87             | 27,5               | 97           | 10,8           |               | 8,33           |
| F8            | 1158          | 75               | 25,83             | 29,9               | 95           | 10,8           |               | 6,48           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 25</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 981           | 19               | 45,78             | 44,91              | 91           | 11,7           | 1,41          | 1,94           |
| F4            | 920           | 15               | 48,53             | 44,65              | 93           | 11,7           | 1,36          | 1,63           |
| F5            | 808           | 52               | 44,69             | 36,11              | 97           | 11,7           | 1,42          | 6,44           |
| F6            | 785           | 72               | 40,93             | 32,13              | 97           | 11,7           | 1,02          | 9,17           |
| F7            | 1012          | 98               | 30,85             | 31,22              | 97           | 10,8           | 1,13          | 9,68           |
| F8            | 1083          | 81               | 33,24             | 36,00              | 96           | 10,8           | 1,33          | 7,48           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 26</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 962           | 29               | 45,78             | 44,04              | 88           | 11,8           |               | 3,01           |
| F4            | 905           | 15               | 48,53             | 43,92              | 90           | 11,8           |               | 1,66           |
| F5            | 756           | 35               | 44,69             | 33,79              | 96           | 11,8           |               | 4,63           |
| F6            | 713           | 28               | 40,93             | 29,18              | 97           | 11,8           |               | 3,93           |
| F7            | 914           | 68               | 30,85             | 28,20              | 97           | 11             |               | 7,44           |
| F8            | 1002          | 79               | 33,24             | 33,31              | 95           | 11             |               | 7,88           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 27</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 933           | 37               | 51,58             | 48,12              | 77           | 12             | 0,92          | 3,97           |
| F4            | 890           | 34               | 53,6              | 47,70              | 80           | 12             | 0,76          | 3,82           |
| F5            | 721           | 18               | 50,9              | 36,70              | 86           | 12             | 1,00          | 2,50           |
| F6            | 685           | 28               | 50,46             | 34,57              | 90           | 12             | 1,61          | 4,09           |
| F7            | 846           | 45               | 34,45             | 29,14              | 97           | 11,2           | 0,85          | 5,32           |

|               |               |                  |                   |                    |              |                |               |                |
|---------------|---------------|------------------|-------------------|--------------------|--------------|----------------|---------------|----------------|
| F8            | 923           | 50               | 36                | 33,23              | 95           | 11,2           | 0,61          | 5,42           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 28</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 896           | 60               | 51,58             | 46,22              | 77,6         | 12,4           |               | 6,70           |
| F4            | 856           | 30               | 53,6              | 45,88              | 78           | 12,4           |               | 3,50           |
| F5            | 703           | 13               | 50,9              | 35,78              | 80           | 12,4           |               | 1,85           |
| F6            | 657           | 26               | 50,46             | 33,15              | 83           | 12,4           |               | 3,96           |
| F7            | 801           | 37               | 34,45             | 27,59              | 98           | 11,2           |               | 4,62           |
| F8            | 873           | 33               | 36                | 31,43              | 98           | 11,2           |               | 3,78           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 29</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 836           | 57               | 59,4              | 49,7               | 102          | 12,3           | 1,01          | 6,82           |
| F4            | 826           | 53               | 66,2              | 54,7               | 108          | 12,3           | 1,51          | 6,42           |
| F5            | 690           | 11               | 58,8              | 40,6               | 108          | 12,3           | 1,03          | 1,59           |
| F6            | 631           | 18               | 56,8              | 35,8               | 112          | 12,3           | 0,85          | 2,85           |
| F7            | 764           | 20               | 40,6              | 31,0               | 93           | 11,2           | 1,17          | 2,62           |
| F8            | 840           | 24               | 41,1              | 34,5               | 94           | 11,2           | 0,95          | 2,86           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 30</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 779           | 43               | 59,4              | 46,27              | 94           | 12,5           |               | 5,52           |
| F4            | 773           | 43               | 66,2              | 51,17              | 97           | 12,5           |               | 5,56           |
| F5            | 679           | 17               | 58,8              | 39,93              | 96           | 12,5           |               | 2,50           |
| F6            | 613           | 19               | 56,8              | 34,82              | 100          | 12,5           |               | 3,10           |
| F7            | 744           | 19               | 40,6              | 30,21              | 99           | 11,3           |               | 2,55           |
| F8            | 816           | 14               | 41,1              | 33,54              | 98           | 11,3           |               | 1,72           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 31</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 736           | 38               | 59,4              | 43,7               |              |                |               | 5,16           |
| F4            | 730           | 40               | 66,2              | 48,3               |              |                |               | 5,48           |
| F5            | 662           | 14               | 58,8              | 38,9               |              |                |               | 2,11           |
| F6            | 594           | 10               | 56,8              | 33,7               |              |                |               | 1,68           |
| F7            | 725           | 8                | 40,6              | 29,4               |              |                |               | 1,10           |
| F8            | 802           | 17               | 46,4              | 37,2               |              |                |               | 2,12           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 32</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 698           | 18               | 78,3              | 54,65              |              |                |               | 2,58           |
| F4            | 690           | 23               | 83                | 57,27              |              |                |               | 3,33           |
| F5            | 648           | 14               | 80                | 51,84              |              |                |               | 2,16           |
| F6            | 584           | 10               | 79,2              | 46,25              |              |                |               | 1,71           |

Can Proteinskimmers improve water quality in recirculation systems so that it effects Growth and Mortality of juvenile Atlantic Halibut (*Hippoglossus hippoglossus*) and can Antibiotic feed help to uncover hidden Bacteria?

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|               |               |                  |                   |                    |              |                |               |                |
|---------------|---------------|------------------|-------------------|--------------------|--------------|----------------|---------------|----------------|
| F7            | 717           | 13               | 56,3              | 40,37              |              |                |               | 1,81           |
| F8            | 785           | 16               | 54                | 42,39              |              |                |               | 2,04           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 33</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 680           | 8                | 79                | 53,7               |              |                | 1,15          | 1,18           |
| F4            | 667           | 17               | 81                | 54,0               |              |                | 0,81          | 2,55           |
| F5            | 634           | 6                | 83                | 52,6               |              |                | 1,39          | 0,95           |
| F6            | 574           | 5                | 80                | 45,9               |              |                | 1,38          | 0,87           |
| F7            | 704           | 8                | 60                | 42,2               |              |                | 1,57          | 1,14           |
| F8            | 769           | 7                | 54                | 41,5               |              |                | 1,1           | 0,91           |
|               |               |                  |                   |                    |              |                |               |                |
| <b>Uke 33</b> | <b>Amount</b> | <b>Mortality</b> | <b>Weight [g]</b> | <b>Biomass[kg]</b> | <b>O2[%]</b> | <b>Temp[C]</b> | <b>SGR(%)</b> | <b>Mort(%)</b> |
| F3            | 672           |                  |                   |                    |              |                |               |                |
| F4            | 650           |                  |                   |                    |              |                |               |                |
| F5            | 628           |                  |                   |                    |              |                |               |                |
| F6            | 569           |                  |                   |                    |              |                |               |                |
| F7            | 696           |                  |                   |                    |              |                |               |                |
| F8            | 762           |                  |                   |                    |              |                |               |                |

| Tanks   | SGR(%) | Mort/day | Mort/week (%) | Mort/month(%) | Akk.Mort (%) | Akk BM dead |
|---------|--------|----------|---------------|---------------|--------------|-------------|
| A+PS    | 1,07   | 0,41     | 2,84          | 11,61         | 43,67        | 20,11       |
| A+PS    | 1,03   | 0,36     | 2,53          | 10,75         | 40,43        | 19,61       |
| PS      | 1,07   | 0,43     | 2,96          | 12,02         | 45,19        | 16,10       |
| PS      | 1,03   | 0,51     | 3,52          | 14,09         | 52,98        | 17,56       |
| Control | 0,95   | 0,51     | 3,49          | 14,06         | 52,86        | 15,99       |
| Control | 0,89   | 0,45     | 3,14          | 12,69         | 47,70        | 16,21       |

|                      | Generell informasjon |              | Status inngående beholdning |               |            | Status utgående beholdning |               |            | Dødelighet  |       | Tilvekst            |                     |                               | Føring                  |                |                     |
|----------------------|----------------------|--------------|-----------------------------|---------------|------------|----------------------------|---------------|------------|-------------|-------|---------------------|---------------------|-------------------------------|-------------------------|----------------|---------------------|
|                      | Enhet                | Utsettnavn   | Antall fisk                 | Snittvekt [g] | Biom. [kg] | Antall fisk                | Snittvekt [g] | Biom. [kg] | Antall fisk | %     | Akk. dødelighet [%] | Netto tilvekst [kg] | Daglig tilvekst i periode [%] | Førfbruk i periode [kg] | Øko. forfaktor | Akk. øko. forfaktor |
| Avdeling: Forsøkskar |                      |              | 6.596                       | 21,70         | 143        | 3.952                      | 72,00         | 284        | 2.644       | 40,08 | 40,08               | 141                 | 1,08                          | 737,60                  | 5,23           | 5,23                |
|                      | F 3                  | Aga Marin 11 | 1.069                       | 22,40         | 24         | 666                        | 79,00         | 53         | 403         | 37,70 | 37,70               | 29                  | 1,13                          | 131,00                  | 4,56           | 4,56                |
|                      | F 4                  | Aga Marin 11 | 997                         | 24,00         | 24         | 651                        | 81,00         | 53         | 346         | 34,70 | 34,70               | 29                  | 1,09                          | 141,70                  | 4,91           | 4,91                |
|                      | F 5                  | Aga Marin 11 | 1.019                       | 23,40         | 24         | 616                        | 83,00         | 51         | 403         | 39,55 | 39,55               | 27                  | 1,14                          | 128,20                  | 4,70           | 4,70                |
|                      | F 6                  | Aga Marin 11 | 1.010                       | 23,70         | 24         | 570                        | 80,00         | 46         | 440         | 43,56 | 43,56               | 22                  | 1,09                          | 123,50                  | 5,69           | 5,69                |
|                      | F 7                  | Aga Marin 11 | 1.232                       | 19,40         | 24         | 689                        | 60,00         | 41         | 543         | 44,07 | 44,07               | 17                  | 1,01                          | 103,10                  | 5,91           | 5,91                |
|                      | F 8                  | Aga Marin 11 | 1.269                       | 18,80         | 24         | 760                        | 54,00         | 41         | 509         | 40,11 | 40,11               | 17                  | 0,95                          | 110,30                  | 6,43           | 6,43                |
|                      |                      |              | 6.596                       | 21,70         | 143        | 3.952                      | 72,00         | 284        | 2.644       | 40,08 | 40,08               | 141                 | 1,08                          | 737,60                  | 5,23           | 5,23                |

Raw data: Water flow, water quality and TGP (%), own measurements:

| Uke 17 | Q[l/min] | V[l] | T[min] | Uke 19 | Q[l/min] | V[l] | T[min] |
|--------|----------|------|--------|--------|----------|------|--------|
| F3     | 42,2     | 1440 | 34,1   | F3     | 42,13    | 1440 | 34,18  |
| F4     | 39,29    | 1440 | 36,7   | F4     | 38,19    | 1440 | 37,71  |
| F5     | 38,75    | 1440 | 37,2   | F5     | 38       | 1440 | 37,89  |
| F6     | 38,73    | 1440 | 37,2   | F6     | 42,13    | 1440 | 34,18  |
| F7     | 36,94    | 1440 | 39,0   | F7     | 32,34    | 1440 | 44,53  |
| F8     | 33,03    | 1440 | 43,6   | F8     | 34,42    | 1440 | 41,84  |
| Uke 24 | TGP(%)   |      |        | Uke 26 | TGP(%)   |      |        |
| F3     | 101,1    |      |        | F3     | 100,9    |      |        |
| F4     | 102,4    |      |        | F4     | 101,5    |      |        |
| F5     | 103      |      |        | F5     | 102,4    |      |        |
| F6     | 102,8    |      |        | F6     | 102,8    |      |        |
| F7     | 100,4    |      |        | F7     | 103,1    |      |        |
| F8     | 100,8    |      |        | F8     | 102,6    |      |        |

From Fishtalk:

| Uke: 19     |          |         |             |      |             |          |         |             |      |
|-------------|----------|---------|-------------|------|-------------|----------|---------|-------------|------|
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato: 10/5  | Sign:    | Stig    |             |      | Dato: 10/5  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          |          |         |             | %    | O2          |          |         |             | %    |
| Temp        | 11,4     |         | 5,8         | °C   | Temp        | 10,7     |         | 6,4         | °C   |
| PH          | 7,2      |         | 7,94        |      | PH          | 7,01     |         | 7,94        |      |
| NH3 NH4+    | 0,05     |         |             | mg/l | NH3 NH4+    | 0,25     |         |             | mg/l |
| NH3         | 0,0004   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0020   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 10,0     |         |             | mg/l | NO3N        | 11       |         |             | mg/l |
| Nitrat      | 44       | 0       | 0           | mg/l | Nitrit      | 48,4     | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 6        |         |             | mg/l | Co2         | 7        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato: 13/5  | Sign:    | Stig    |             |      | Dato: 13/5  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 99       |         | 95          | %    | O2          | 97       |         | 95          | %    |
| Temp        | 11,5     |         | 5,8         | °C   | Temp        | 10,9     |         | 6,4         | °C   |
| PH          | 7,18     |         | 7,94        |      | PH          | 7,02     |         | 7,94        |      |
| NH3 NH4+    | 0,1      |         |             | mg/l | NH3 NH4+    | 0,25     |         |             | mg/l |
| NH3         | 0,0008   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0020   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 9,0      |         |             | mg/l | NO3N        | 11       |         |             | mg/l |
| Nitrat      | 39,6     | 0       | 0           | mg/l | Nitrit      | 48,4     | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 7        |         |             | mg/l | Co2         | 8        |         |             | mg/l |

| Uke: 21     |          |         |             |      |             |          |         |             |      |
|-------------|----------|---------|-------------|------|-------------|----------|---------|-------------|------|
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato:24/5   | Sign:    | Stig    |             |      | Dato: 24/5  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 99       |         | 97          | %    | O2          | 98       |         | 97          | %    |
| Temp        | 11,4     |         | 5,8         | °C   | Temp        | 10,8     |         | 6,4         | °C   |
| PH          | 7,15     |         | 7,93        |      | PH          | 7,02     |         | 7,93        |      |
| NH3 NH4+    | 0,1      |         |             | mg/l | NH3 NH4+    | 0,25     |         |             | mg/l |
| NH3         | 0,0008   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0020   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 10,0     |         |             | mg/l | NO3N        | 10       |         |             | mg/l |
| Nitrat      | 44       | 0       | 0           | mg/l | Nitrit      | 44       | 0       | 0           | mg/l |
| NO2N        | 0,05     |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0,165    | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 5        |         |             | mg/l | Co2         | 7        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |
| Uke: 22     |          |         |             |      |             |          |         |             |      |
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato:31/5   | Sign:    | Stig    |             |      | Dato: 31/5  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 101      |         | 96          | %    | O2          | 97       |         | 96          | %    |
| Temp        | 11,3     |         | 5,9         | °C   | Temp        | 10,2     |         | 5,9         | °C   |
| PH          | 7,25     |         | 7,98        |      | PH          | 7,03     |         | 7,98        |      |
| NH3 NH4+    | 0,25     |         |             | mg/l | NH3 NH4+    | 0,5      |         |             | mg/l |
| NH3         | 0,0020   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0040   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 10,0     |         |             | mg/l | NO3N        | 11       |         |             | mg/l |
| Nitrat      | 44       | 0       | 0           | mg/l | Nitrit      | 48,4     | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 7        |         |             | mg/l | Co2         | 7        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |

| Uke: 23     |          |         |             |      |             |          |         |             |  |
|-------------|----------|---------|-------------|------|-------------|----------|---------|-------------|--|
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |  |
| Dato: 7/6   | Sign:    | Stig    |             |      | Dato: 7/6   | Sign:    | Stig    |             |  |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |  |
| O2          | 101      |         | 96          | %    | O2          | 97       |         | 96          |  |
| Temp        | 10,6     |         | 6,2         | °C   | Temp        | 11,7     |         | 6,2         |  |
| PH          | 7,22     |         | 8,02        |      | PH          | 7,04     |         | 8,02        |  |
| NH3 NH4+    | 0,1      |         |             | mg/l | NH3 NH4+    | 0,5      |         |             |  |
| NH3         | 0,0008   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0040   | 0,0000  | 0,0000      |  |
| NO3N        | 8,0      |         |             | mg/l | NO3N        | 10       |         |             |  |
| Nitrat      | 35,2     | 0       | 0           | mg/l | Nitrit      | 44       | 0       | 0           |  |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             |  |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           |  |
| Co2         | 7        |         |             | mg/l | Co2         | 7        |         |             |  |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             |  |
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |  |
| Dato: 10/6  | Sign:    | Stig    |             |      | Dato: 10/6  | Sign:    | Stig    |             |  |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |  |
| O2          | 104      |         | 98          | %    | O2          | 102      |         | 98          |  |
| Temp        | 12,1     |         | 6,4         | °C   | Temp        | 11,2     |         | 6,4         |  |
| PH          | 7,27     |         | 7,94        |      | PH          | 7,02     |         | 7,94        |  |
| NH3 NH4+    | 0        |         |             | mg/l | NH3 NH4+    | 0,25     |         |             |  |
| NH3         | 0,0000   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0020   | 0,0000  | 0,0000      |  |
| NO3N        | 8,0      |         |             | mg/l | NO3N        | 11       |         |             |  |
| Nitrat      | 35,2     | 0       | 0           | mg/l | Nitrit      | 48,4     | 0       | 0           |  |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             |  |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           |  |
| Co2         | 5        |         |             | mg/l | Co2         | 7        |         |             |  |

| Uke: 24     |          |         |             |      |             |          |         |             |      |
|-------------|----------|---------|-------------|------|-------------|----------|---------|-------------|------|
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato: 14/6  | Sign:    | Stig    |             |      | Dato: 14/6  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 101      |         | 97          | %    | O2          | 101      |         | 97          | %    |
| Temp        | 11,9     |         | 6,5         | °C   | Temp        | 11       |         | 6,5         | °C   |
| PH          | 7,27     |         | 7,96        |      | PH          | 7,02     |         | 7,96        |      |
| NH3 NH4+    | 0,05     |         |             | mg/l | NH3 NH4+    | 0,2      |         |             | mg/l |
| NH3         | 0,0004   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0016   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 10,0     |         |             | mg/l | NO3N        | 10       |         |             | mg/l |
| Nitrat      | 44       | 0       | 0           | mg/l | Nitrit      | 44       | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 5        |         |             | mg/l | Co2         | 6        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato: 17/6  | Sign:    | Stig    |             |      | Dato: 17/6  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 102      |         |             | %    | O2          | 99       |         |             | %    |
| Temp        | 11,9     |         |             | °C   | Temp        | 11       |         |             | °C   |
| PH          | 7,25     |         |             |      | PH          | 7        |         |             |      |
| NH3 NH4+    | 0,05     |         |             | mg/l | NH3 NH4+    | 0,5      |         |             | mg/l |
| NH3         | 0,0004   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0040   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 6,0      |         |             | mg/l | NO3N        | 10       |         |             | mg/l |
| Nitrat      | 26,4     | 0       | 0           | mg/l | Nitrit      | 44       | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 6        |         |             | mg/l | Co2         | 6        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |



*Can Proteinskimmers improve water quality in recirculation systems so that it effects Growth and Mortality of juvenile Atlantic Halibut (Hippoglossus hippoglossus) and can Antibiotic feed help to uncover hidden Bacteria?*

2011

| Uke: 25     |          |         |             |      |             |          |         |             |      |
|-------------|----------|---------|-------------|------|-------------|----------|---------|-------------|------|
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato: 21/6  | Sign:    | Stig    |             |      | Dato: 21/6  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 100      |         | 96          | %    | O2          | 98       |         | 96          | %    |
| Temp        | 12       |         | 6,5         | °C   | Temp        | 11,1     |         | 6,5         | °C   |
| PH          | 7,25     |         | 8           |      | PH          | 7,01     |         | 8           |      |
| NH3 NH4+    | 0        |         |             | mg/l | NH3 NH4+    | 0,2      |         |             | mg/l |
| NH3         | 0,0000   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0016   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 10,0     |         |             | mg/l | NO3N        | 10       |         |             | mg/l |
| Nitrat      | 44       | 0       | 0           | mg/l | Nitrit      | 44       | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 6        |         |             | mg/l | Co2         | 7        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato: 24/6  | Sign:    | Stig    |             |      | Dato: 24/6  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 98       |         | 95          | %    | O2          | 98       |         | 95          | %    |
| Temp        | 11,5     |         | 6,2         | °C   | Temp        | 10,7     |         | 6,2         | °C   |
| PH          | 7,24     |         | 7,95        |      | PH          | 7,02     |         | 7,95        |      |
| NH3 NH4+    | 0,1      |         |             | mg/l | NH3 NH4+    | 0,1      |         |             | mg/l |
| NH3         | 0,0008   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0008   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 8,0      |         |             | mg/l | NO3N        | 10       |         |             | mg/l |
| Nitrat      | 35,2     | 0       | 0           | mg/l | Nitrit      | 44       | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0        |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0        | 0       | 0           | mg/l |
| Co2         | 6        |         |             | mg/l | Co2         | 7        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |

| Uke: 26     |          |         |             |      |             |          |         |             |      |
|-------------|----------|---------|-------------|------|-------------|----------|---------|-------------|------|
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato: 28/6  | Sign:    | Stig    |             |      | Dato: 28/6  | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 102      |         | 95          | %    | O2          | 102      |         | 95          | %    |
| Temp        | 11,6     |         | 6,5         | °C   | Temp        | 10,8     |         | 6,5         | °C   |
| PH          | 7,14     |         | 7,95        |      | PH          | 7        |         | 7,95        |      |
| NH3 NH4+    | 0,1      |         |             | mg/l | NH3 NH4+    | 0,1      |         |             | mg/l |
| NH3         | 0,0008   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0008   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 8,0      |         |             | mg/l | NO3N        | 10       |         |             | mg/l |
| Nitrat      | 35,2     | 0       | 0           | mg/l | Nitrit      | 44       | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0        |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0        | 0       | 0           | mg/l |
| Co2         | 6        |         |             | mg/l | Co2         | 7        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato: 1/7   | Sign:    | Stig    |             |      | Dato: 1/7   | Sign:    | Stig    |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 99       |         | 93          | %    | O2          | 98       |         | 93          | %    |
| Temp        | 11,9     |         | 6,6         | °C   | Temp        | 11       |         | 6,6         | °C   |
| PH          | 7,13     |         | 7,93        |      | PH          | 7,01     |         | 7,93        |      |
| NH3 NH4+    | 0,25     |         |             | mg/l | NH3 NH4+    | 0,15     |         |             | mg/l |
| NH3         | 0,0020   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0012   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 9,0      |         |             | mg/l | NO3N        | 4        |         |             | mg/l |
| Nitrat      | 39,6     | 0       | 0           | mg/l | Nitrit      | 17,6     | 0       | 0           | mg/l |
| NO2N        | 0        |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0        | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 6        |         |             | mg/l | Co2         | 7        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |

*Can Proteinskimmers improve water quality in recirculation systems so that it effects Growth and Mortality of juvenile Atlantic Halibut (Hippoglossus hippoglossus) and can Antibiotic feed help to uncover hidden Bacteria?*

2011

| Uke: 29     |          |         |             |      |             |          |         |             |      |
|-------------|----------|---------|-------------|------|-------------|----------|---------|-------------|------|
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato:19/7   | Sign:    | Trude   |             |      | Dato: 16/7  | Sign:    | Trude   |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 95       |         | 91          | %    | O2          | 98       |         | 91          | %    |
| Temp        | 12,4     |         | 6,8         | °C   | Temp        | 11,2     |         | 6,8         | °C   |
| PH          | 7,09     |         | 7,96        |      | PH          | 7,15     |         | 7,96        |      |
| NH3 NH4+    | 0,5      |         |             | mg/l | NH3 NH4+    | 0,5      |         |             | mg/l |
| NH3         | 0,0040   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0040   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 10,0     |         |             | mg/l | NO3N        | 6        |         |             | mg/l |
| Nitrat      | 44       | 0       | 0           | mg/l | Nitrit      | 26,4     | 0       | 0           | mg/l |
| NO2N        | 0,05     |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0,165    | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 8        |         |             | mg/l | Co2         | 7        |         |             | mg/l |
| Alkalinitet |          |         |             | mg/l | Alkalinitet |          |         |             | mg/l |
| A - HALL    |          |         |             |      | B - HALL    |          |         |             |      |
| Dato:22/7   | Sign:    | Trude   |             |      | Dato:22/7   | Sign:    | Trude   |             |      |
| A-Hall      | Nivåtank | Kar nr: | Inntaksvann |      | B-Hall      | Nivåtank | Kar nr: | Inntaksvann |      |
| O2          | 98       |         | 88          | %    | O2          | 99       |         | 88          | %    |
| Temp        | 12,3     |         | 6,8         | °C   | Temp        | 11,1     |         | 6,8         | °C   |
| PH          | 7,15     |         | 7,97        |      | PH          | 7,15     |         | 7,97        |      |
| NH3 NH4+    | 0,25     |         |             | mg/l | NH3 NH4+    | 0,25     |         |             | mg/l |
| NH3         | 0,0020   | 0,0000  | 0,0000      | mg/l | NH3         | 0,0020   | 0,0000  | 0,0000      | mg/l |
| NO3N        | 8,0      |         |             | mg/l | NO3N        | 6        |         |             | mg/l |
| Nitrat      | 35,2     | 0       | 0           | mg/l | Nitrit      | 26,4     | 0       | 0           | mg/l |
| NO2N        | 0,05     |         |             | mg/l | NO2N        | 0,05     |         |             | mg/l |
| Nitrit      | 0,165    | 0       | 0           | mg/l | Nitrat      | 0,165    | 0       | 0           | mg/l |
| Co2         | 7        |         |             | mg/l | Co2         | 7        |         |             | mg/l |

*Can Proteinskimmers improve water quality in recirculation systems so that it effects Growth and Mortality of juvenile Atlantic Halibut (Hippoglossus hippoglossus) and can Antibiotic feed help to uncover hidden Bacteria?*

2011

Data Fiskefarm Raunes

| Uke: 33+34 | Antall | Snittvekt(g) | Biomasse (kg) | Døde | Formengde (kg)   | Tilvekst (%) | Dødelighet(%) | FCR | Tetthet(kg/m2) |
|------------|--------|--------------|---------------|------|------------------|--------------|---------------|-----|----------------|
| Kar 5      | 3637   | 37,62        | 136,82        | 57   |                  | 1,36         | 1,57          |     | 4,83           |
| Kar 6      | 6074   | 19,48        | 118,32        | 63   |                  | 1,50         | 1,04          |     | 4,18           |
| Kar 7      |        |              | 0,00          |      |                  | #DIV/0!      | #DIV/0!       |     | 0,00           |
| Kar 8      | 35793  | 10,3         | 368,67        | 688  |                  | 1,51         | 1,92          |     | 7,33           |
| Kar 9      |        |              |               |      |                  |              |               |     |                |
| Uke: 35+36 | Antall | Snittvekt(g) | Biomasse (kg) | Døde | Formengde (kg/d) | Tilvekst (%) | Dødelighet(%) | FCR | Tetthet(kg/m2) |
| Kar 5      | 14179  | 39,93        | 566,17        | 229  | 8,09             | 1,43         | 1,62          |     | 20,01          |
| Kar 6      | 6011   | 27,5         | 165,30        | 14   | 3,44             | 2,08         | 0,23          |     | 5,84           |
| Kar 7      | 0      | 0,00         | 0,00          | 0    | #DIV/0!          | #DIV/0!      | #DIV/0!       |     | 0,00           |
| Kar 8      | 35105  | 15,28        | 536,40        | 205  | -1,74            | -0,32        | 0,58          |     | 10,66          |
| Kar 9      | 1820   | 401          | 729,82        | 1    | 0,05             | 0,01         | 0,05          |     | 14,51          |
| Uke: 37+38 | Antall | Snittvekt(g) | Biomasse (kg) | Døde | Formengde (kg/d) | Tilvekst (%) | Dødelighet(%) | FCR | Tetthet(kg/m2) |
| Kar 5      | 13884  | 48,7         | 676,15        | 54   | 10,14            | 0,53         | 0,39          |     | 23,89          |
| Kar 6      | 6218   | 36,7         | 228,20        | 22   | 3,42             | 0,13         | 0,35          |     | 8,06           |
| Kar 7      |        | 10           | 0,00          | 73   | 0,00             | -100,00      | #DIV/0!       |     | 0,00           |
| Kar 8      | 34900  | 14,6         | 509,54        | 363  | 7,64             | 2,91         | 1,04          |     | 10,13          |
| Kar 9      | 1813   | 401,4        | 727,74        | 0    | 4,00             | 4,71         | 0             |     | 14,47          |
| Uke: 39+40 | Antall | Snittvekt(g) | Biomasse (kg) | Døde | Formengde (kg/d) | Tilvekst (%) | Dødelighet(%) | FCR | Tetthet(kg/m2) |
| Kar 5      | 13830  | 52,7         | 728,8         | 48   | 13,12            | 0,53         | 0,35          |     | 25,75          |
| Kar 6      | 6196   | 37,4         | 231,7         | 21   | 4,17             | 0,13         | 0,34          |     | 8,19           |
| Kar 7      | 2000   |              | 0,0           | 56   | 0,00             | -100,00      | 2,8           |     | 0,00           |
| Kar 8      | 34537  | 22,44        | 775,0         | 38   | 13,95            | 2,91         | 0,11          |     | 15,41          |
| Kar 9      | 1813   | 800          | 1450,4        | 4    | 5,00             | 4,71         | 0,22          |     | 28,83          |
| Uke 41-42  | Antall | Snittvekt(g) | Biomasse (kg) | Døde | Formengde (kg)   | Tilvekst (%) | Dødelighet(%) | FCR | Tetthet(kg/m2) |
| Kar 5      | 13662  | 66,3         | 905,8         | 35   | 13,970           | 1,54         | 0,26          |     | 18,01          |
| Kar 6      | 6295   | 44,2         | 278,2         | 8    | 3,116            | 1,12         | 0,13          |     | 9,83           |
| Kar 7      | 1944   | 10           | 19,4          | 45   | #DIV/0!          | #DIV/0!      | 2,31          |     | 0,39           |
| Kar 8      | 34499  | 25,6         | 883,2         | 118  | 15,897           | 0,88         | 0,34          |     | 17,56          |
| Kar 9      | 1809   | 600          | 1085,4        | 0    | 5,427            | 0,50         | 0,00          |     | 21,58          |
| Uke: 43-44 | Antall | Snittvekt(g) | Biomasse (kg) | Døde | Formengde (kg)   | Tilvekst (%) | Dødelighet(%) | FCR | Tetthet(kg/m2) |
| Kar 5      | 1899   | 14           | 26,6          | 21   | 0,647            | 2,43         | 1,11          |     | 0,94           |
| Kar 6      | 6287   | 51,3         | 322,5         | 12   | 3,450            | 1,07         | 0,19          |     | 11,44          |
| Kar 7      | 13627  | 88,06        | 1200,0        | 23   | 24,576           | 2,05         | 0,17          |     | 23,86          |
| Kar 8      | 34381  | 30,08        | 1034,2        | 58   | 11,982           | 1,16         | 0,17          |     | 20,56          |
| Kar 9      | 1809   | 600          | 1085,4        | 4    | 0,000            | 0,00         | 0,22          |     | 21,58          |
| Uke: 45-46 | Antall | Snittvekt(g) | Biomasse (kg) | Døde | Formengde (kg)   | Tilvekst (%) | Dødelighet(%) | FCR | Tetthet(kg/m2) |
| Kar 5      | 1878   | 16           | 30,0          | 54   | 0,3              | 0,96         | 2,88          |     | 0,60           |
| Kar 6      | 6275   | 77,02        | 483,3         | 15   | 14,2             | 2,95         | 0,24          |     | 9,61           |
| Kar 7      | 13604  | 112,6        | 1531,8        | 29   | 27,1             | 1,77         | 0,21          |     | 30,45          |
| Kar 8      | 34323  | 37,27        | 1279,2        | 72   | 19,7             | 1,54         | 0,21          |     | 25,43          |
| Kar 9      | 1805   | 600          | 1083,0        | 0    | 0,0              | 0,00         | 0,00          |     | 21,53          |
| Uke: 47-48 | Antall | Snittvekt(g) | Biomasse (kg) | Døde | Formengde (kg)   | Tilvekst (%) | Dødelighet(%) | FCR | Tetthet(kg/m2) |
| Kar 5      | 1824   | 20           | 36,5          | 40   | 0,6              | 1,61         | 2,19          |     | 0,73           |
| Kar 6      | 6260   | 81,79        | 512,0         | 8    | 2,2              | 0,43         | 0,13          |     | 10,18          |
| Kar 7      | 13575  | 140,78       | 1911,1        | 7    | 30,7             | 1,61         | 0,05          |     | 37,99          |
| Kar 8      | 34251  | 53,65        | 1837,6        | 96   | 48,4             | 2,64         | 0,28          |     | 36,53          |
| Kar 9      | 1805   | 601          | 1084,8        | 0    | 0,1              | 0,01         | 0             |     | 21,57          |

Data-Farmcontroll:

| <u>Generation</u> | <u>Opphav</u>   | <u>Kar</u> | <u>Period</u> | <u>Antall</u> | <u>Snittvekt</u> | <u>Biomasse</u> | <u>Dødelighet</u> | <u>Døde [%]</u> |
|-------------------|-----------------|------------|---------------|---------------|------------------|-----------------|-------------------|-----------------|
| 2004              | Marine Harvest  | A1         | 01.12.2005    | 2498          | 1103             | 2756            | 0                 | 0               |
|                   | Marine Harvest  |            | 19.07.2005    | 2374          | 1460             | 3467            | 124               | 4,69            |
| 2004              | Marine Harvest  | A2         | 01.12.2005    | 4795          | 507              | 2424            | 0                 | 0               |
|                   | Marine Harvest  |            | 19.07.2005    | 3285          | 967              | 3177            | 267               | 7,52            |
| 2004              | Marine Harvest  | A4         | 01.12.2005    | 2561          | 800              | 2050            | 0                 | 0               |
|                   | Marine Harvest  |            | 19.07.2005    | 2444          | 1442             | 3524            | 117               | 4,6             |
| 2004              | Marine Harvest  | A5         | 01.12.2005    | 2917          | 550              | 1604            | 0                 | 0               |
|                   | Marine Harvest  |            | 19.07.2005    | 2656          | 1111             | 2951            | 261               | 8,9             |
| 2005              | Aga             | A7         | 02.12.2005    | 12737         | 24               | 306             | 0                 | 0               |
|                   | Aga             |            | 06.04.2006    | 10313         | 96               | 990             | 2424              | 19,03           |
| 2005              | Aga             | A8         | 10.12.2005    | 11648         | 52               | 605             | 8                 | 0,07            |
|                   | Aga             |            | 07.04.2006    | 10411         | 153              | 1583            | 1245              | 10,68           |
| 2006              | Brandal Harvest | A9         | 01.01.2006    | 7769          | 20               | 155             | 0                 | 0               |
|                   | Brandal Harvest |            | 01.12.2005    | 7143          | 73               | 521             | 626               | 8               |
| 2006              | Brandal Harvest | A10        | 25.01.2006    | 8099          | 34               | 275             | 0                 | 0               |
|                   | Brandal Harvest |            | 11.04.2006    | 7452          | 90               | 671             | 647               | 7,99            |
| 2005              | Aga             | A18        | 15.12.2005    | 11843         | 21               | 243             | 0                 | 0               |
|                   | Aga             |            | 05.04.2006    | 8378          | 78               | 656             | 3465              | 29,26           |
| 2005              | Aga             | A19        | 15.12.2005    | 12346         | 17               | 214             | 0                 | 0               |
| 2005              | Aga             |            | 05.04.2005    | 8544          | 75               | 641             | 3802              | 30,8            |
| 2005              | Aga             | A20        | 15.12.2005    | 12565         | 12               | 155             | 0                 | 0               |
| 2005              | Aga             |            | 05.04.2005    | 9420          | 58               | 546             | 3145              | 25,03           |
| 2005              | Fiskey          | B9         | 14.02.2006    | 8357          | 17               | 142             | 0                 | 0               |
| 2005              | Fiskey          |            | 05.03.2006    | 8341          | 24               | 200             | 16                | 0,19            |
| 2005              | Fiskey          | B10        | 14.02.2006    | 8000          | 17               | 136             | 0                 | 0               |
| 2005              | Fiskey          |            | 05.03.2006    | 7954          | 24,66            | 195,8           | 55                | 0,69            |
| 2006              | Fiskey          | B10        | 14.02.2006    | 16282         | 28,01            | 456             | 0                 | 0               |
| 2006              | Fiskey          |            | 13.04.2006    | 15152         | 39               | 590             | 1134              | 6,96            |
| 2006              | Fiskey          | A8         | 07.10.2006    | 8600          | 25               | 215             | 0                 | 0               |
| 2006              | Fiskey          |            | 02.01.2007    | 5800          | 67               | 388             | 2800              | 32,56           |
| 2005              | Fiskey          | A9         | 07.10.2006    | 11196         | 25               | 280             | 0                 | 0               |
| 2005              | Fiskey          |            | 02.01.2007    | 6948          | 57               | 395             | 4248              | 37,94           |
| 2005              | Sande seafarm   | A10        | 18.09.2006    | 4565          | 123              | 562             | 0                 | 0               |
| 2005              | Sande seafarm   |            | 06.03.2007    | 4337          | 519              | 2251            | 1224              | 4,99            |
| 2006              | Sande seafarm   | A20        | 18.09.2006    | 6035          | 99               | 596             | 0                 | 0               |
| 2006              | Sande seafarm   |            | 06.03.2007    | 5515          | 427              | 2355            | 520               | 8,62            |
| 2006              | Sande seafarm   | B10        | 30.07.2006    | 5400          | 100              | 540             | 0                 | 0               |

|      |                 |     |            |       |     |      |      |       |
|------|-----------------|-----|------------|-------|-----|------|------|-------|
| 2006 | Sande seafarm   |     | 24.09.2006 | 4176  | 159 | 666  | 1224 | 22,67 |
| 2006 | Sande seafarm   | B20 | 30.07.2006 | 5400  | 100 | 540  | 0    | 0     |
| 2006 | Sande seafarm   |     | 24.09.2006 | 4634  | 171 | 793  | 766  | 14,19 |
| 2006 | Aga             | A5  | 27.02.2007 | 9144  | 33  | 302  | 0    | 0     |
| 2006 | Aga             |     | 03.10.2007 | 7951  | 75  | 592  | 1193 | 13    |
| 2006 | Aga             | A7  | 27.02.2007 | 3229  | 53  | 172  | 0    | 0     |
| 2006 | Aga             |     | 10.05.2007 | 2689  | 111 | 299  | 540  | 16,72 |
| 2006 | Aga             | A15 | 27.02.2007 | 7333  | 54  | 396  | 0    | 0     |
| 2006 | Aga             |     | 03.10.2007 | 6610  | 103 | 681  | 723  | 9,8   |
| 2006 | Sande seafarm   | A6  | 01.07.2007 | 6329  | 107 | 676  | 0    | 0     |
| 2006 | Sande seafarm   |     | 20.08.2007 | 6071  | 160 | 976  | 258  | 4     |
| 2006 | Sande seafarm   | A7  | 01.07.2007 | 3305  | 142 | 469  | 0    | 0     |
| 2006 | Sande seafarm   |     | 20.08.2007 | 3166  | 187 | 592  | 139  | 4,21  |
| 2006 | Aga             | C4  | 23.04.2007 | 5052  | 45  | 226  | 0    | 0     |
| 2006 | Aga             |     | 17.09.2007 | 4029  | 154 | 620  | 431  | 9,7   |
| 2006 | Aga             | C7  | 23.04.2007 | 8112  | 38  | 311  | 0    | 0     |
| 2006 | Aga             |     | 06.06.2007 | 7322  | 45  | 327  | 35   | 0,48  |
| 2006 | Aga             | C8  | 23.04.2007 | 8710  | 33  | 290  | 0    | 0     |
| 2006 | Aga             |     | 17.09.2007 | 7132  | 89  | 635  | 847  | 10,62 |
| 2007 | Aga             | C3  | 15.05.2007 | 1789  | 46  | 83   | 0    | 0     |
| 2007 | Aga             |     | 17.09.2007 | 1617  | 99  | 160  | 180  | 10,02 |
| 2006 | Brandal Harvest | A11 | 18.04.2007 | 4900  | 25  | 124  | 0    | 0     |
| 2006 | Brandal Harvest |     | 02.10.2007 | 4444  | 152 | 675  | 456  | 9,31  |
| 2006 | Aga/Fjord H     | C1  | 20.07.2007 | 9465  | 25  | 238  | 0    | 0     |
| 2006 | Aga/Fjord H     |     | 18.09.2007 | 8166  | 48  | 392  | 1299 | 13,72 |
| 2006 | Aga/Fjord H     | C2  | 20.07.2007 | 6478  | 37  | 239  | 0    | 0     |
| 2006 | Aga/Fjord H     |     | 18.09.2007 | 5720  | 77  | 440  | 758  | 11,76 |
| 2006 | Fiskey          | A6  | 01.02.2007 | 7706  | 45  | 347  | 0    | 0     |
| 2006 | Fiskey          |     | 24.04.2007 | 6305  | 97  | 610  | 1401 | 16,87 |
| 2006 | Aga             | A16 | 01.02.2007 | 1569  | 45  | 71   | 0    | 0     |
| 2006 | Aga             |     | 24.04.2007 | 1252  | 96  | 120  | 317  | 20,2  |
| 2006 | Fiskey          | A17 | 01.02.2007 | 6694  | 45  | 301  | 0    | 0     |
| 2006 | Fiskey          |     | 24.04.2007 | 5534  | 106 | 586  | 1160 | 17,67 |
| 2006 | Fiskey          | A17 | 08.01.2007 | 14263 | 36  | 512  | 0    | 0     |
| 2006 | Fiskey          |     | 15.05.2007 | 10718 | 128 | 1372 | 3194 | 22,39 |
| 2007 | Sande seafarm   | A10 | 25.11.2007 | 5314  | 180 | 957  | 0    | 0     |
| 2007 | Sande seafarm   |     | 09.04.2008 | 5103  | 464 | 2369 | 211  | 3,97  |
| 2007 | Brandal Harvest | A20 | 14.12.2007 | 5500  | 23  | 129  | 0    | 0     |
| 2007 | Brandal Harvest |     | 10.06.2008 | 5110  | 184 | 943  | 390  | 7,09  |
| 2007 | Brandal         | A19 | 14.12.2007 | 11600 | 19  | 223  | 0    | 0     |

|             |                        |            |                   |              |              |             |             |              |
|-------------|------------------------|------------|-------------------|--------------|--------------|-------------|-------------|--------------|
|             | <u>Harvest</u>         |            |                   |              |              |             |             |              |
| <u>2007</u> | <u>Brandal Harvest</u> |            | <u>10.06.2008</u> | <u>9517</u>  | <u>114</u>   | <u>1137</u> | <u>1640</u> | <u>14,14</u> |
| <u>2007</u> | <u>Aga/Fjord H</u>     | <u>A1</u>  | <u>22.01.2008</u> | <u>12504</u> | <u>22</u>    | <u>271</u>  | <u>0</u>    | <u>0</u>     |
| <u>2007</u> | <u>Aga/Fjord H</u>     |            | <u>05.06.2008</u> | <u>9695</u>  | <u>69</u>    | <u>672</u>  | <u>2809</u> | <u>22,46</u> |
| <u>2007</u> | <u>Aga/Fjord H</u>     | <u>A2</u>  | <u>22.01.2008</u> | <u>9560</u>  | <u>28</u>    | <u>265</u>  | <u>0</u>    | <u>0</u>     |
| <u>2007</u> | <u>Aga/Fjord H</u>     |            | <u>05.06.2008</u> | <u>7575</u>  | <u>160</u>   | <u>1175</u> | <u>1985</u> | <u>20,76</u> |
| <u>2007</u> | <u>Aga/Fjord H</u>     | <u>A11</u> | <u>22.01.2008</u> | <u>22887</u> | <u>16</u>    | <u>335</u>  | <u>0</u>    | <u>0</u>     |
| <u>2007</u> | <u>Aga/Fjord H</u>     |            | <u>08.04.2008</u> | <u>20892</u> | <u>36</u>    | <u>756</u>  | <u>1995</u> | <u>8,72</u>  |
| <u>2007</u> | <u>Brandal Harvest</u> | <u>A12</u> | <u>18.03.2008</u> | <u>13040</u> | <u>25</u>    | <u>327</u>  | <u>0</u>    | <u>0</u>     |
| <u>2007</u> | <u>Brandal Harvest</u> |            | <u>12.07.2008</u> | <u>11280</u> | <u>86</u>    | <u>972</u>  | <u>1760</u> | <u>13,5</u>  |
| <u>2008</u> | <u>Scotia Halibut</u>  | <u>A6</u>  | <u>30.09.2008</u> | <u>8502</u>  | <u>78</u>    | <u>668</u>  | <u>0</u>    | <u>0</u>     |
| <u>2008</u> | <u>Scotia Halibut</u>  |            | <u>08.02.2009</u> | <u>7520</u>  | <u>133</u>   | <u>1005</u> | <u>1370</u> | <u>15,4</u>  |
| <u>2007</u> | <u>Scotia Halibut</u>  | <u>A12</u> | <u>27.10.2008</u> | <u>7344</u>  | <u>75</u>    | <u>550</u>  | <u>0</u>    | <u>0</u>     |
| <u>2007</u> | <u>Scotia Halibut</u>  |            |                   | <u>6588</u>  | <u>114</u>   | <u>751</u>  | <u>756</u>  | <u>10,3</u>  |
| <u>2008</u> | <u>Scotia Halibut</u>  | <u>A13</u> | <u>30.09.2008</u> | <u>4663</u>  | <u>47</u>    | <u>219</u>  | <u>0</u>    | <u>0</u>     |
| <u>2008</u> | <u>Scotia Halibut</u>  |            | <u>08.02.2009</u> | <u>3099</u>  | <u>91</u>    | <u>282</u>  | <u>1564</u> | <u>33,5</u>  |
| <u>2008</u> | <u>Scotia Halibut</u>  | <u>A10</u> | <u>11.12.2008</u> | <u>10390</u> | <u>13</u>    | <u>135</u>  | <u>0</u>    | <u>0</u>     |
| <u>2008</u> | <u>Scotia Halibut</u>  |            | <u>01.04.2009</u> | <u>7764</u>  | <u>44</u>    | <u>343</u>  | <u>2626</u> | <u>25</u>    |
| <u>2008</u> | <u>Aga</u>             | <u>B8</u>  | <u>03.02.2009</u> | <u>18166</u> | <u>9,94</u>  | <u>180</u>  | <u>0</u>    | <u>0</u>     |
| <u>2008</u> | <u>Aga</u>             |            | <u>05.06.2009</u> | <u>11840</u> | <u>38</u>    | <u>382</u>  | <u>6276</u> | <u>34,64</u> |
| <u>2008</u> | <u>Scotia Halibut</u>  | <u>B19</u> | <u>03.02.2009</u> | <u>17063</u> | <u>25</u>    | <u>427</u>  | <u>0</u>    | <u>0</u>     |
| <u>2008</u> | <u>Scotia Halibut</u>  |            | <u>05.06.2009</u> | <u>12695</u> | <u>101</u>   | <u>1287</u> | <u>4368</u> | <u>25,6</u>  |
| <u>2008</u> | <u>Risör</u>           | <u>B20</u> | <u>03.02.2009</u> | <u>10082</u> | <u>22,7</u>  | <u>229</u>  | <u>0</u>    | <u>0</u>     |
| <u>2008</u> | <u>Risör</u>           |            | <u>05.06.2009</u> | <u>8915</u>  | <u>101</u>   | <u>903</u>  | <u>1167</u> | <u>11,5</u>  |
| <u>2009</u> | <u>Aga G09</u>         | <u>A9</u>  | <u>04.12.2009</u> | <u>13458</u> | <u>13,5</u>  | <u>818</u>  | <u>0</u>    | <u>0</u>     |
| <u>2009</u> | <u>Aga G09</u>         |            | <u>08.03.2010</u> | <u>7841</u>  | <u>31,33</u> | <u>245</u>  | <u>5617</u> | <u>41,86</u> |
| <u>2009</u> | <u>Aga G09</u>         | <u>A10</u> | <u>04.12.2009</u> | <u>8912</u>  | <u>19,5</u>  | <u>173</u>  | <u>0</u>    | <u>0</u>     |
| <u>2009</u> | <u>Aga G09</u>         |            | <u>08.03.2010</u> | <u>5799</u>  | <u>43,83</u> | <u>254</u>  | <u>2042</u> | <u>25</u>    |
| <u>2009</u> | <u>Aga G09</u>         | <u>A20</u> | <u>04.12.2009</u> | <u>9372</u>  | <u>14,5</u>  | <u>135</u>  | <u>0</u>    | <u>0</u>     |
| <u>2009</u> | <u>Aga G09</u>         |            | <u>08.03.2010</u> | <u>5331</u>  | <u>34,7</u>  | <u>185</u>  | <u>3996</u> | <u>37,08</u> |
| <u>2009</u> | <u>Risör</u>           | <u>A8</u>  | <u>11.03.2010</u> | <u>15450</u> | <u>39</u>    | <u>602</u>  | <u>0</u>    | <u>0</u>     |
| <u>2009</u> | <u>Risör</u>           |            | <u>01.07.2010</u> | <u>13794</u> | <u>105</u>   | <u>1451</u> | <u>1656</u> | <u>10,7</u>  |
| <u>2009</u> | <u>Risör</u>           | <u>A19</u> | <u>11.03.2010</u> | <u>10308</u> | <u>49</u>    | <u>505</u>  | <u>0</u>    | <u>0</u>     |
| <u>2009</u> | <u>Risör</u>           |            | <u>01.07.2010</u> | <u>9101</u>  | <u>121</u>   | <u>1105</u> | <u>1207</u> | <u>11,7</u>  |
| <u>2009</u> | <u>Aga G09</u>         | <u>A14</u> | <u>29.06.2010</u> | <u>10193</u> | <u>25</u>    | <u>257</u>  | <u>0</u>    | <u>0</u>     |
| <u>2009</u> | <u>Aga G09</u>         |            | <u>01.10.2010</u> | <u>4754</u>  | <u>100</u>   | <u>478</u>  | <u>373</u>  | <u>7,28</u>  |
| <u>2009</u> | <u>Aga G09</u>         | <u>A15</u> | <u>29.06.2010</u> | <u>10610</u> | <u>25</u>    | <u>268</u>  | <u>0</u>    | <u>0</u>     |
| <u>2009</u> | <u>Aga G09</u>         |            | <u>01.10.2010</u> | <u>4809</u>  | <u>89</u>    | <u>429</u>  | <u>5801</u> | <u>54,6</u>  |
| <u>2009</u> | <u>Risör</u>           | <u>A4</u>  | <u>28.07.2010</u> | <u>11587</u> | <u>42</u>    | <u>487</u>  | <u>4</u>    | <u>0</u>     |
| <u>2009</u> | <u>Risör</u>           |            | <u>07.09.2010</u> | <u>10663</u> | <u>65</u>    | <u>693</u>  | <u>928</u>  | <u>8,01</u>  |
| <u>2009</u> | <u>Risör</u>           | <u>A5</u>  | <u>28.07.2010</u> | <u>7755</u>  | <u>43</u>    | <u>334</u>  | <u>6</u>    | <u>0</u>     |

*Can Proteinskimmers improve water quality in recirculation systems so that it effects Growth and Mortality of juvenile Atlantic Halibut (Hippoglossus hippoglossus) and can Antibiotic feed help to uncover hidden Bacteria?*

**2011**

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|             |              |  |                   |             |           |            |            |            |
|-------------|--------------|--|-------------------|-------------|-----------|------------|------------|------------|
| <u>2009</u> | <u>Risör</u> |  | <u>07.09.2010</u> | <u>7030</u> | <u>65</u> | <u>454</u> | <u>731</u> | <u>9,4</u> |
|-------------|--------------|--|-------------------|-------------|-----------|------------|------------|------------|