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Behavioral Experiments for One-Way Trafficking and Individual Monitoring of Atlantic Salmon, Salmo Salar.

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Aquaculture

Acknowledgement

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Sigrid Venås

Abstract

The following experiments were set up to imitate the behavior of entering a feeding station, enabling individual monitoring of feed intake in Atlantic salmon, *Salmon salar*. This will not only increase the feed efficiency directly, but also be of great interest regarding breeding, enabling more accurate research of the heritability of feed efficiency traits in Atlantis salmon. In the first experiment hatches for one-way trafficking of fish were tested, by dividing a tank into two sides by a plexiglass wall attaching one-way hatches to two holes in the wall in different directions, having the fish swim through a hatch to access feed in one direction only. Inspired by traditional fish traps and fishing gear the hatches were cone-formed with the exit pointing out from the wall, limiting the fish's ability to detect the exit opening, and cross through in the wrong direction. Further, the direction of the current through the hatches and the hatch size were tested to see how it would affect the cross behavior. The fish was monitored by a web-camera installed over the tank, programmed to take pictures every second during meals and record when movement through the hatches were detected. This led to thousands of pictures and hours of videos used to observe the cross behavior. Close to 100% of all crosses through the hatches were in the right direction and the fish preferred to cross against the current. The shape of the hatch was of greater importance than the direction of the current through the hatches. Further, three experiments were done to investigate the possibilities of getting one fish to enter a feeding area through one hatch, and back into a holding area through another hatch, and whether this behavior could be affected by increased flow and different feeding-regimens. Plexiglass were used to install a restricted feeding area in the tank, with the hatches used in the first experiment attached to each side to secure crossing in one direction only. The ability to utilize the feeding area increased over time and with higher flow through the hatches. These experiments demonstrate the possibilities of Atlantic salmon to enter a restricted area to feed, and the behavior observed can be used to further develop feeding stations for individual monitoring in aquaculture.

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1. Introduction

In Norway, the value of contribution from the Norwegian seafood industry was NOK 94 billion in 2017, where aquaculture activity accounted for about NOK 62 billion. The costs in the Norwegian aquaculture industry rose 50% from 2012 to 2016, where both the sea lice problem and the rising of fish feed cost was the main reasons (Richardsen et al., 2017). From 2014 to 2016 the feed cost increased from NOK 14 a kilo of slaughtered Atlantic salmon, *Salmo salar*, to NOK 18 a kilo, which is due to both increased feed prices and higher use of feed per kilo produced (Richardsen et al., 2017). Today, most feeding control methods in aquaculture is based on calculations and monitoring of larger stocks and groups (Zhou et al., 2018). Finding a method making it possible to obtain accurate records of individual feed intake in Atlantic salmon formed the basis of my thesis. The aim of the project is to develop a feeding station that makes it possible to feed and monitor salmon individually, going from group feeding to precision feeding, improving the feed efficiency in the aquaculture industry. Not only will this reduce pollution from the aquaculture industry, it also makes it possible to study the traits for feed utilization. The idea is to create a closed station where the fish can enter to feed, leave when finished, having the feed spill collected in the station, resulting in accurate individual data on feed intake.

In Norway, the breeding program of Atlantic salmon started in the 1970s, where an increased growth rate, higher age of sexual maturation and better disease resistance, flesh color and fat content has been important breeding goals (Gjerde et al., 2007). In a study assessing whether selection for increased growth rate in Atlantic salmon is associated with increased feed intake and/or better feed utilization, showed that fish of the selected line had a significantly lower intake of protein and energy per kilogram (Thodesen et al., 1999). In another study by Thodesen et al. (2001), the results agreed with those presented in 1999. This study concluded that feed consumption, growth and feed utilization in Atlantic salmon may be improved by selective breeding. The study also showed that feed utilization can be improved by selection for increased growth, but the response may decline as the growth rate is increased (Thodesen et al., 2001). Improving feed efficiency in fish using selective breeding has been showed to be difficult due to the environment they live in and group sizes they are held in (De Verdal et al., 2018). Today the heritability estimates of feed efficiency traits in fish is assumed to be relatively low, which might be a result inaccurate measurements on feed intake (De Verdal et al., 2018). Individual monitoring of feeding might increase the chances of studying these traits further.

Precision Fish Farming (PFF) aims to apply control-engineering principles to fish production, improving the famer’s ability to monitor, control and document biological processes in fish farms (Føre et al., 2018). Compared to Precision Livestock Farming (PLF), aquaculture farming faces additional challenges complicating the PFF (Føre et al., 2018). Not only does the number of individual animals in fish farms exceeds what is common in most terrestrial livestock farms, the feeding are also enacted on the entire cage population and not on individuals or small group levels (Føre et al., 2018). Today most feeding strategies in salmon production is based on feeding tables suggesting feed amounts depending on population size and temperature, where the ongoing conflict between good growth rates and feed spillage in conjunction with over- and underfeeding is a daily struggle that has consequences for both fish welfare and farm economy (Føre et al., 2018). The agricultural concept of precision feeding relies on the between-animal variation, involving the use of feeding techniques that allow not only the right amount of feed, but also the right composition to be provided at the right time for each individual (Pomar et al., 2011). The systems today are based feeding stations. When an individual enters the feeding station, and recognizes the individuals by a sensor that monitors each individuals unique electronic tag (Frost et al., 1997). After entering the feeding station the individual can be weighed and the amount of feed consumed can be measured (Frost et al., 1997). The key to the solution is to understand fish behavior and learning. Fish behavior is a result of a combination of learned association to extrinsic stimuli and intrinsic drives, such as hunger and stress (Fernö et al., 2011). The intrinsic and extrinsic factors affect the fish motivation and influence the attention directed to relevant stimuli, and again the willingness to explore the environment where this stimuli occur (Warburton, 2003).

The experiments done in this study is a continuation of three behavioral experiments of Atlantic Salmon for individualized feeding stations presented in a master thesis by Matthew Chernin (2019). The experiments were set out to test whether Atlantic Salmon would pass through a narrow hatch, imitating the action of entering a feeding station, and whether this behavior could be influenced (Chernin, 2019). An upward trend in crossing through the hatches occurred as the time in the experiment increased, in addition the experiment showed a preference of crossing the opening against the current (Chernin, 2019). To further develop the feeding station, the idea of getting the fish to cross through the feeding station from one side and out on the other side occurred. To achieve this one-way trafficking of fish a hatch was designed, inspired by traditional fish-trap (Collins, 1990). Most traditional fish traps are designed to have the fish enter through a hole for bait, not being able to find the opening and as a result being trapped (Gabriel et al., 2008). In an experiment done by Collins et. Al (1990), three different trap designs were tested, and the ‘chevron’ design was the most effective. The hatch in the ‘chevron’ trap was cone-formed, having the fish swim in through the wider opening, follow the edge to the narrow end, and be trapped in the holding area of the trap (Collins, 1990). This was the inspiration for the one-way hatches used in these experiments. During the first experiment three different set-ups was tested to understand how the current through the hatch and the hatch size would affect the fish behavior. Further, three experiments were done to investigate the possibilities of getting one fish to enter a feeding area through one hatch, and back into a holding area through another hatch, and whether this behavior could be affected of increased flow and different feeding-regimens.

2. Material and method

2.1 Experiment 1

2.1.1 Materials

The experiment was conducted from November 1st to December 13th, 2018, at the center for fish research at NMBU, Campus Ås. As a continuation of the experiments done by Matthew Chernin (2919) the same 9 Atlantic Salmon, was used in the experiment. The average fish weight was 231,3 ±10,9 grams and the average length was 26,4 ± 0,6 cm. The average fish height was 6,28 ± 0,26 cm and the average width was 3,83 ± 0,25 cm. The experiment was divided into three parts; exp 1.1 – testing of one-way hatches with countercurrent through the hatches (01.11.2018-15.11.2018), exp 1.2 - testing of one-way hatches with cocurrent through the hatches (15.11.2018-29.11.2018) and exp 1.3 - Testing of smaller one-way hatches with countercurrent through the hatches (29.11.2018-13.12.2018).

The hatch design was inspired by traditional fish traps where the aim was to have the fish go into the trap, but not get out. The hatches in such traps are often cone-formed, having the fish go in through the wider side of the hatch, follow the wall and go through the narrow side of the hatch and into the trap. The narrow side of the hatch points out in the open rom in the trap, making it hard to find. The hatches made for the experiments were cone-formed, made from see-through 7mm thick plexiglass (figure 1).

The tank used in the experiment was a round with a diameter of 1,4 m and a water height of 1,0 m (1300 L). The tank was divided into two sides, side 1 and side 2, using a see through 7 mm thick plexiglass wall. The hatches were placed at each side of the tank 20 cm from the tank circumference and 45 cm from the bottom of the tank (figure 2). The tank was supplied with water from two inlet pipes on each side of the tank to secure equal water supplies on both sides of the tank, and a circular water current during the experiments. The water flow on each side was 15 l/min during all experiments. The two inlet pipes had three holes, with a diameter of 7 mm, vertically on the lower end of the pipe, directing the water flow along the walls of the tank (figure 1). The water temperature was 15 degrees Celsius. Two belt feeders were installed on each side of the tank, both controlled with a timer at the socket to schedule the meals (figure 2). Skretting Nutra Olympic 3mm pellets were used throughout the experiment. Because of the feeders attached to the rim of the tank, no lid was used. To prevent the fish from jumping out of the tank, a half meter high jump guard was attached to the rim. The ceiling lights in the experiment room were kept on throughout the day.

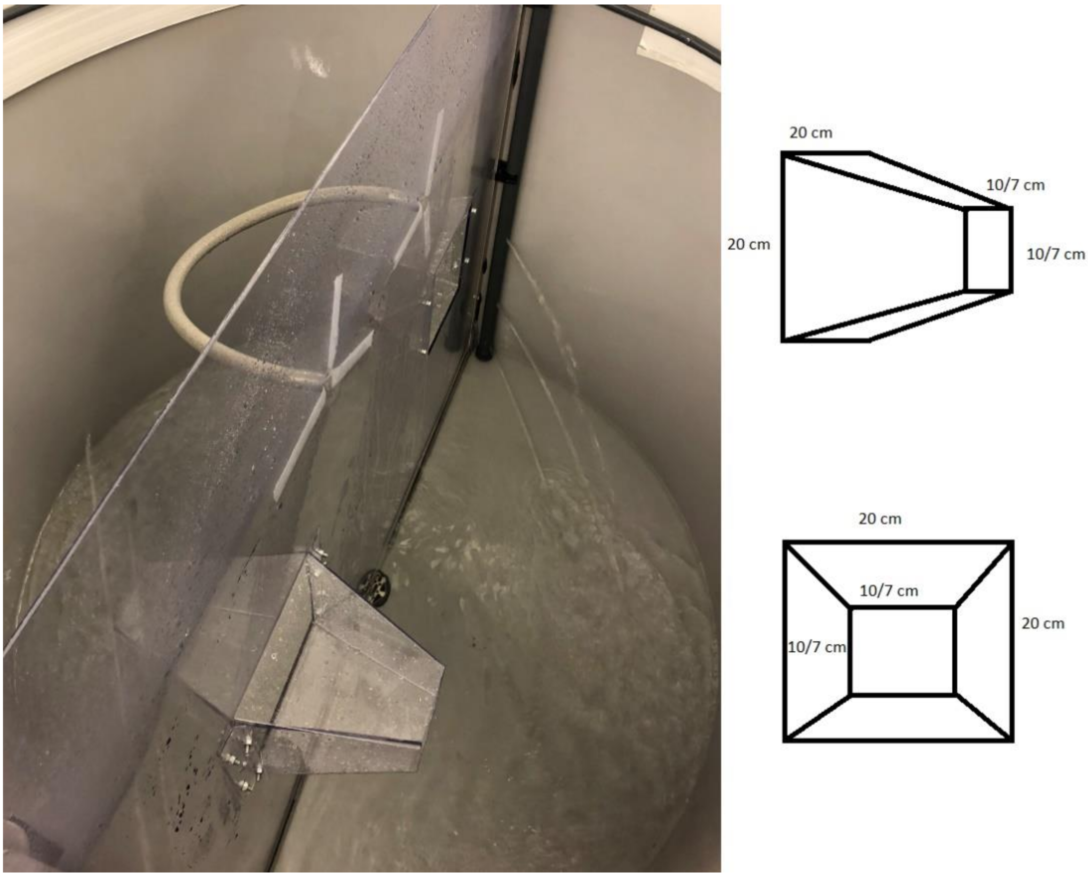


*Figure 1: Tank set-up with the plexiglass wall dividing the tank into two sides, with attached hatches to facilitate one-way trafficking of fish. The hatch-design used in all experiments, with the exit opening variating between 10cm*10cm and 7cm*7cm testing different sizes. The inlet pipes with the vertically holes directing the water flow along the walls of the tank.*

2.1.1.1 Experiment 1.1

In exp.1.1 the hatches with a 20cm*20cm entrance and a 10cm*10cm exit was tested (figure 1), with countercurrent through the hatches created by the set water current in the tank (figure 2). The aim of the set-up was to see if it was possible to create a one-way traffic of fish through the hatches, making the fish pass over from side one to side two through hatch 2, and back to side one through hatch 1.

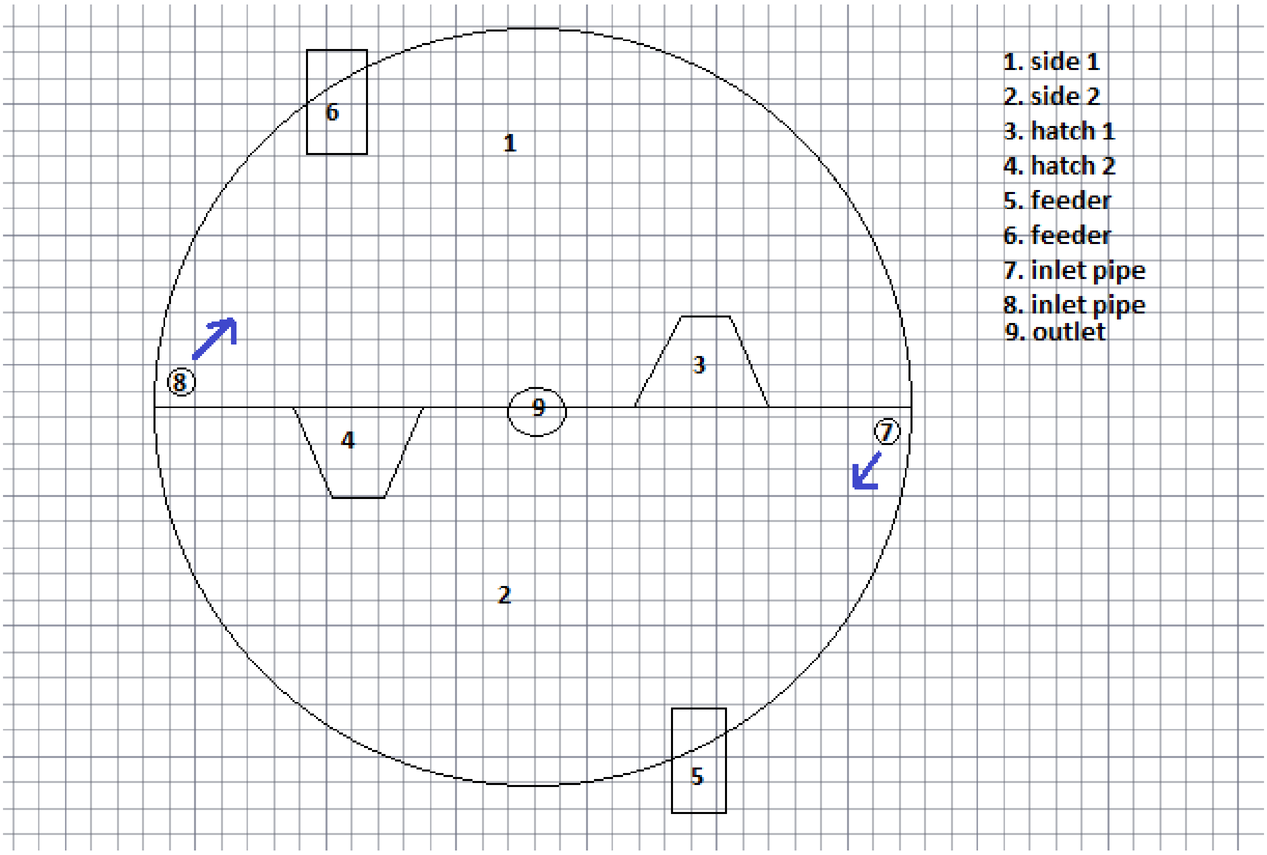


*Figure 2: Tank set up in experiment 1.1 and 1.3 with the plexiglass wall dividing the tank into two sides and attached hatches for one-way trafficking of fish. The hatch size was 10 cm*10 cm and in exp.1.1 and 7 cm * 7 cm in exp.1.3 to test the effect of different hatch sizes. The direction of the inlet pipes created countercurrent through the hatches.*

2.1.1.2 Experiment 1.2

The average fish weight and length at the start of the experiment was 257,0 ± 12,3 grams and 27,5 ± 0,65 cm. During experiment 1.2 the same hatches as used in experiment 1.1 was set up in the opposite direction, creating cocurrent through the hatches (figure 3), to see how it would affect the fish behavior.

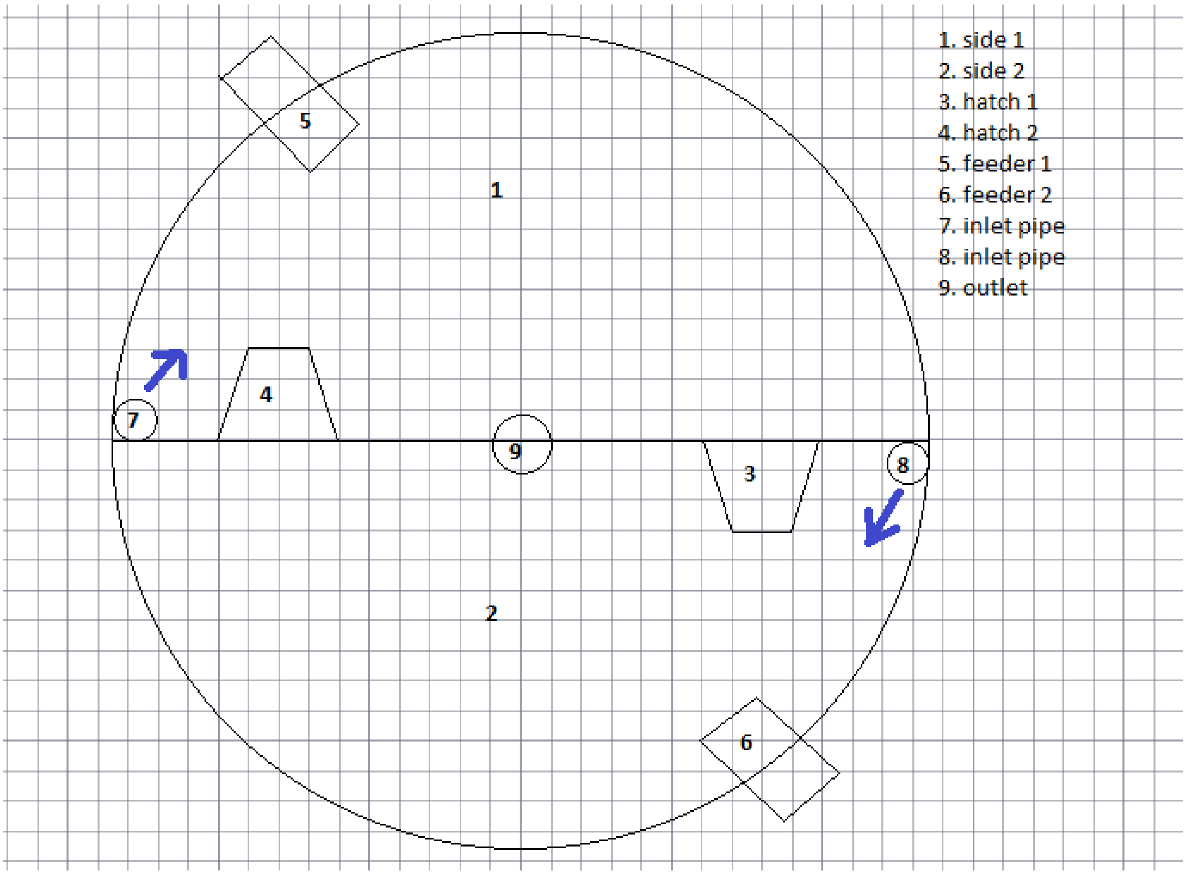


*Figure 3: Tank set up experiment 1.2 with the inlet pipes crating cocurrent through the hatches to test the effect on the cross behavior. Hatch size 10 cm * 10 cm.*

2.1.1.3 Experiment 1.3

The average fish weight and length at the start of the experiment was 296,4 ± 15,8 grams and 28,4 ± 0,73 cm. The fish had an average height of 6,5 ± 0,06 cm and an average width of 3,83 ± 0,03 cm. Using the same set-up as in experiment 1.1, smaller hatch was tested to see if it would affect the cross behavior, with an opening of 20cm*20cm and an exit of 7cm*7cm (figure 2), which leaves a gap of 0,5 cm compared to the fish height.

2.1.2 Method

The fish were individually weighed, and measurements of length, height and width were taken before introduced to the tank on side one. The fish were sedated using Tricaine Methanesulfonate, MS-222, when weighed and measured. They were given two meals per day, one in the evening on side one, and one in the morning on side two, each meal lasting for 30 minutes, making a 11.5 hours period between each meal. The first meal after weigh-in was given on the same side where the fish were introduced. During the experiment the fish were weighed and length-measured once a week before introduced to the tank on side one. A web-camera was installed overlooking the whole tank. Using a program called FishSpy, the camera was scheduled to take 1 picture per second during the morning and evening meal. The rest of the day, motion detection was used, where the camera was programmed to record when motion through the hatches were detected. The pictures and videos from the monitoring were gone through daily, observing and collecting data from the cross behavior.

2.2 Experiment 2

2.2.1 Materials

The experiment was conducted from January 17th to February 7th, 2019, at the center for fish research at NMBU, Campus Ås. The same 9 Atlantic Salmon, from exp.1 was also used in the following experiment. The average fish weight was 387 ± 39,9 grams and the average length was 31,4 ± 1,2 cm at the start of the experiment.

The same tank that was used in experiment one was also used in experiment two. The wall dividing the tank into two sides were removed, and a separated feeding area was made of two see-through plexiglass walls in a 70 degrees’ angle from the center of the tank, making a feeding area in 1/5th of the tank and a holding area in 4/5th of the tank (figure 4). The wider hatches (10 cm*10cm) from exp.1 was placed in opposite directions on each wall. Hatch one was attached approximately 25 cm over the tank bottom and 20 cm from the rim and hatch two, approximately 45 cm over the tank bottom and 20 cm from the rim. The inlet pipes were placed the same way as in exp.1, with a water flow of 15 l/min on each side. A belt-feeder was installed over the feeding area, controlled with a timer at the socket to schedule the meals. The aim of the set-up was to see if the fish would go into a restricted feeding area through a hatch, and back out to the holding area of the tank through another hatch, in one direction only.

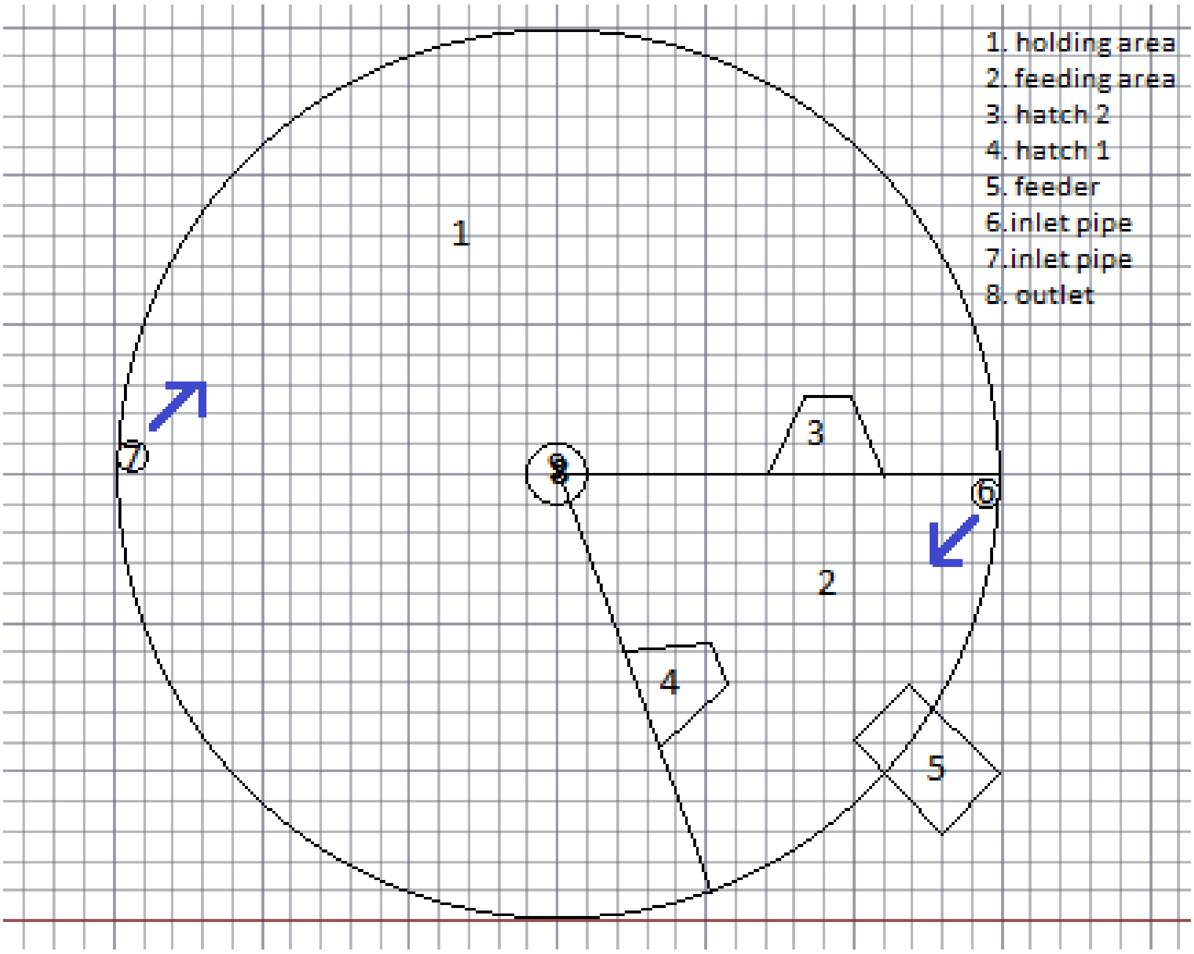


Figure 4: Tank set-up during experiment two, three and four, with a restricted feeding area made of plexiglass, and one-way hatches to secure one-way traffic through the feeding area.

2.2.2 Method

The fish was individually weighed and length-measured before being introduced in the holding area of the tank January 17th, 2019. The experiment was running for 3 weeks and the fish was weighed and length-measured the first week (January 24th) an at the end of the experiment (February 7th). This was to see if we would get better growth with no handling for two weeks. The same feeding routine as in experiment one was used, with two meals per day from 09:00-09:30 and 21:00-21:30, both meals were fed in the feeding area. The fish was monitored the same way as in the first experiment with pictures during the meals and motion detection during the rest of the day.

2.3 Experiment 3 – Materials and methods

The experiment was conducted from February 7th to February 19th, 2019, at the center for fish research at NMBU, Campus Ås. The 9 Atlantic Salmon from experiment two was also used in experiment three. The average fish weight was 379±31,8 grams and the average length was 31,8±1,5 cm at the start of the experiment.

In this experiment continuously feeding throughout the day was tested to see if the fish would respond differently when food was always available in the feeding area. The belt-feeder was programmed to run for 2 seconds, then paused for 16 seconds to feed as close to continuously as possible. The fish was individually weighed and length-measured before introduced to the holding area of the tank. The experiment was running for 2 weeks and the fish was weighed and length-measured once a week. The fish was monitored by using motion detection during the whole day.

2.4 Experiment 4 – Material and method

The experiment was conducted from February 19th to March 19th, 2019, at the center for fish research at NMBU, Campus Ås. The 9 Atlantic Salmon from experiment three was also used in experiment four. The average fish weight was 383±41,4 grams and the average length was 32±1,3 cm at the start of the experiment.

During experiment three the fish growth and the number of crosses was lower than expected, which gave a suspicion of bad flow though the hatches leading to low stimuli of the fish. An experiment was done get an overview of the water flow in the feeding area and through the hatches. Food-color were pumped into the inlet pipe in the feeding area, to get a picture of the flow in the tank (figure 5). There were no feed-color flowing through the hatches, meaning that there was no or very low flow through the hatches. Most of the color were sucked out of the tank through the outlet in the bottom of the tank.

Figure 5: Food-color test. Food-color were pumped into the inlet pipe in the feeding area to demonstrate the flow through the hatches. The lack of color streaming out of hatch one, indicates a low flow though the hatch.

To see if better flow through the hatches would lead to better growth, two small pumps were installed to create better flow through the hatches. From the pumps, two tubes were attached to the exit side of the hatches (the narrow side), creating countercurrent through the hatches (figure 6). The same feeder that was used in experiment 3 was also used in experiment 4, with continuously feeding throughout the day. The fish was individually weighed and length-measured before set-out outside the feeding-area of the tank 19.02.2019. The experiment was running for 4 weeks and the fish was weighed and length-measured once a week. The fish was monitored by using motion detection during the whole day.

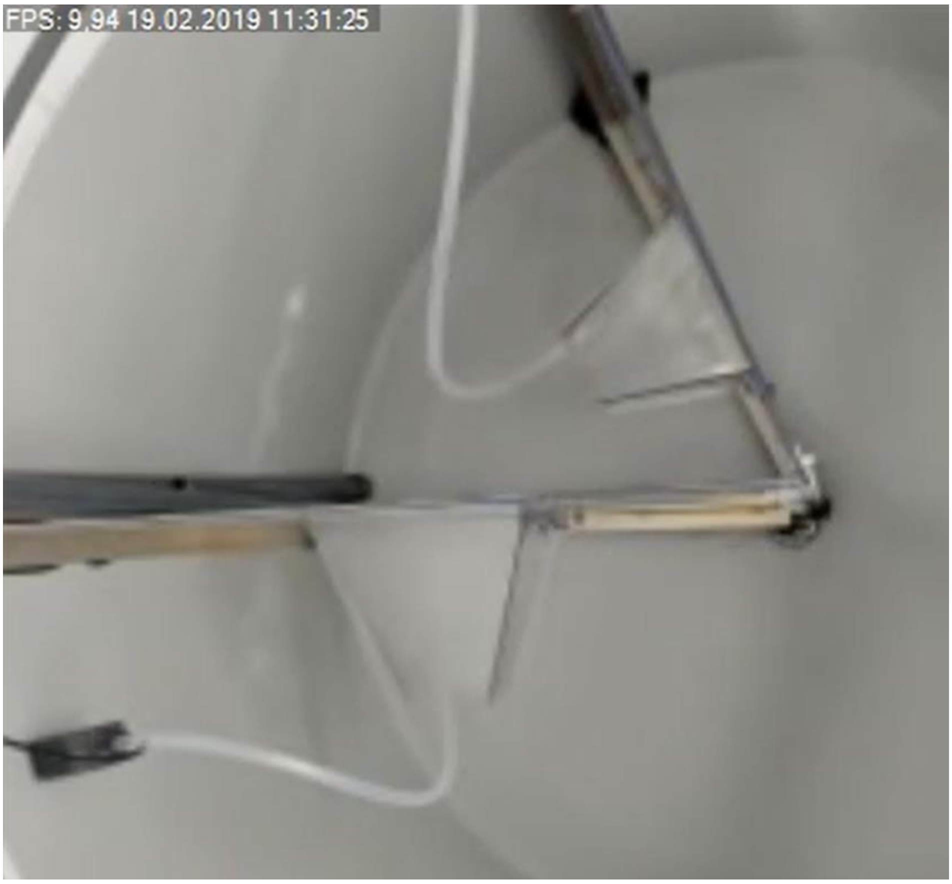


Figure 6: Feeding area with pumps attached to the tank wall, with tubes leading to the hatches exit pumping water through the hatches, increasing the flow.

2.5 Data collection

The data were collected during the experiment period from November 2018 to March 2019 and was based on the pictures and videos from the web camera monitoring. A high definition Logitech web camera was installed overlooking the whole tank and monitoring the fish throughout the day. Using a program called Scorpion Caption, the camera was programmed and scheduled. The data collection was divided into two parts, were the two first experiments distinguished between meals and the rest of the day, and the last two had continuously feeding throughout the day. During the meals the camera was programmed to take one picture per second, resulting in 2400 pictures per meal. During the non-meal periods of the day motion detection was used, recording when motion through the hatches were detected. The motion detection gave a lot of false positives because it also recorded when the fish were moving over and under the hatches. This gave a total of 2400 pictures per meal, meaning 4800 pictures per day, and often hundreds of short videos from the motion detection resulted in hours of work per day going through the pictures and videos, registering the crosses and observing the behavior.

The day was divided into four phases: morning long series (from 09:40 PM to 09:00 AM), morning meal (09:00 am to 09:40 am), evening long series (09:40 am to 09:00 pm) and evening meal (09:00 pm to 09:40pm). Using the pictures and videos from the monitoring, the fish behavior during meals and during non-meals was recorded. Both right-way (from the wider side of the hatch to the narrow side) and wrong-way (from the narrow side of the hatch to the wider side) crossings with their time, and the number of fishes on each side of the wall, was recorded. In experiment two, three and four the number of fishes inside the feeding area throughout the day was recorded by counting the number of fishes inside the feeding area at each video. These registrations were used to calculate the total number of crossings each day and crossings per minute both during meals and non-meals. The collected data was analyzed using linear regression where the slopes and r-squared values were compared to determine their significance (Løvås, 2013). In addition, the data was portioned into weekly summaries to compare the different weeks of the experiments. To analyze the variability in the cross behavior and growth standard deviation and coefficient of variation were used (Løvås, 2013). In addition, a timeline analyses were done to look for any increase in the fishes crossing behavior the last two hours before meal, compared to the rest of the day.

Growth statistic for the fish were also collected and calculated. The weights and lengths of each fish were registered at the start of each experiment, and once a week during the experiments. These data were used to calculate the weekly average weight and length, and the standard deviation and variance for each week. These data were used to compare the weekly trends in growth during the experiments. In addition the data were used to calculate the Specific Growth Rate (SGR), the Feed Conversion Ratio (FCR) and the condition factor (Fulton, 1902).

Formula 1: Specific Growth Rate.

$$\boldsymbol{SGR = \frac{\ln(B_1) - \ln(B_0)}{t}}$$

B_1 = Final biomass

B_0 = Starting biomass

t = number of days

SGR = Daily Specific Growth Rate

Formula 2: Feed Conversion Ratio.

$$\boldsymbol{FCR = \frac{Total\ feed\ given}{B_1 - B_0}}$$

B_1 = Final biomass

B_0 = Starting biomass

FCR = Feed Conversion Rate

Formula 3: Condition Factor

$$\boldsymbol{K = \frac{W}{L^3} * 100}$$

W = Weight in grams

L = Fork Length in Centimeters

K = Condition Factor

3. Results

3.1 Total crosses per day from the master thesis by, Matt Chernin (2019)

The results from Matthew Chernin's master thesis (2019), shows an average number of crosses per day at 180,71 during the first week and 199,29 during the second week (table 1), meaning an increase in the number of crosses per day over time.

Table 1: Weekly summary of total number of crosses and average number of crosses per day, from the results from Matthew Cherning's master thesis (2019).

Weekly summary: Total Crosses and Average Crosses per Day				
Period	**Average Crosses per Day**	**Total crosses**	**Standard Deviation**	**Variance**
Week 1 Days 1-7	180,71	1265	18,55	343,92
Week 2 Days 8-14	199,29	1395	15,4	237,06

3.2 Experiment 1 – testing one-way hatches

3.2.1 Experiment .1.1 – week 1-2

3.2.1.1 Total crosses per day

Total number of crosses decreased during the two weeks' period (figure 7). The linear regression analysis gives a slope of minus 11,03, which means an average decrease of 11,03 crosses per day of the experiment. The regression analysis also suggests a variation of 33,6 % that can be explained by the model. The total number of crosses per day variated greatly from 337 on day 1, to 63 on day 5, and 37 before take-out on day 14 (figure 7). When doing the regression analysis for week 2 only, the R-square value of the trendline is 0,76, suggesting that a variation of 76% can be explained by the model.

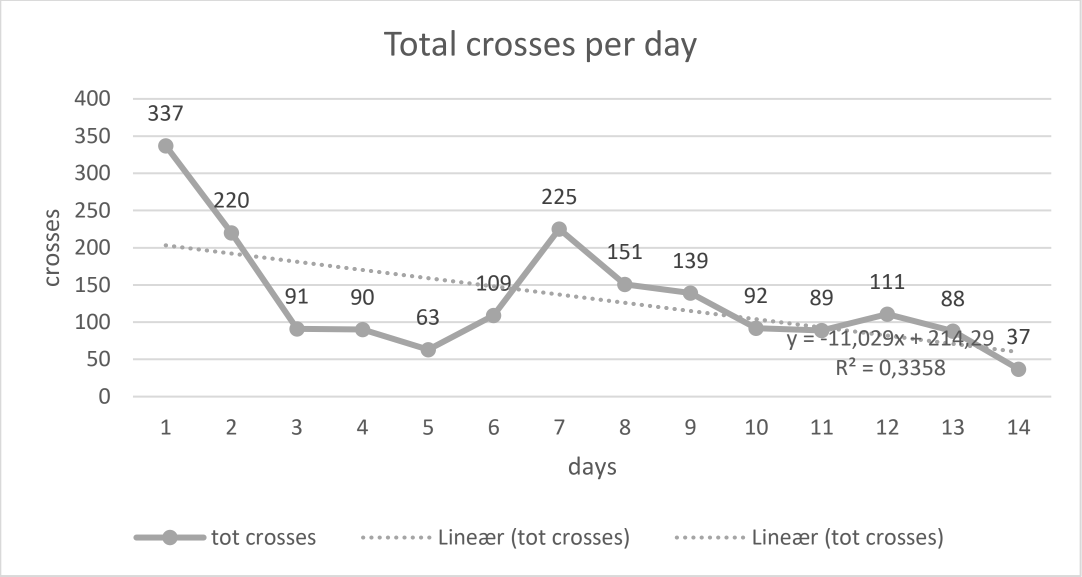


Figure 7: Total crosses per day through both hatches from experiment 1.1 with a regression analysis showing a decrease in the total number of crosses per day.

The results show an increase in the average crosses per week, with an average of 162 crosses per day in week 1 and 101 crosses per day in week 2 (table 2). The standard deviation and variation did decrease during the two weeks period, suggesting a more consistent cross behavior over time (table 1). The greatest variation is seen in week 1 (table 2), which also contains the day of the highest number of crosses (figure 7).

Table 2: Weekly summary with average crosses per day, total crosses per week, standard deviation and variance from experiment 1.1.

Week #	Average crosses per day	Total crosses	Standard deviation	Variance
1	162	1133	100,66	10132,14
2	101	932	37,71	1422,33

3.2.1.2 Cross direction through the hatches

Close to 100 % of all crosses were countercurrent, from the wider side to the narrow side, with 99,8 % in week 1 and 99,7 % in week 2 (table 3). During week 1 only two crosses in the wrong direction was registered (table 3), and both crosses appeared on the first day of the experiment during the evening meal, which also was the day with the greatest number of crosses (figure 7). The three wrong way crosses during week 2 appeared on day 12 and 13, with respectively one and two crosses, were two of them appeared during the evening meal.

Table 3: Percentage of right way crosses through the hatches for week one and two in experiment 1.1.

Week #	Right way	Wrong way	Total	Right way crosses (%)
1	969	2	971	99,8
2	874	3	877	99,7

3.2.1.3 Crosses per minute during meal and non-meal periods

The total number of crosses were further divided into the number of crosses per period, and the average crosses per minute during meals and non-meals were calculated (figure 8). The negative slopes of the linear regression trendlines for the periods is in consistent with the decrease of total crosses over time (figure 8). The results show an increase of cross activity when feed is available and the crosses during meal is the greatest contributor to the total number of crosses.

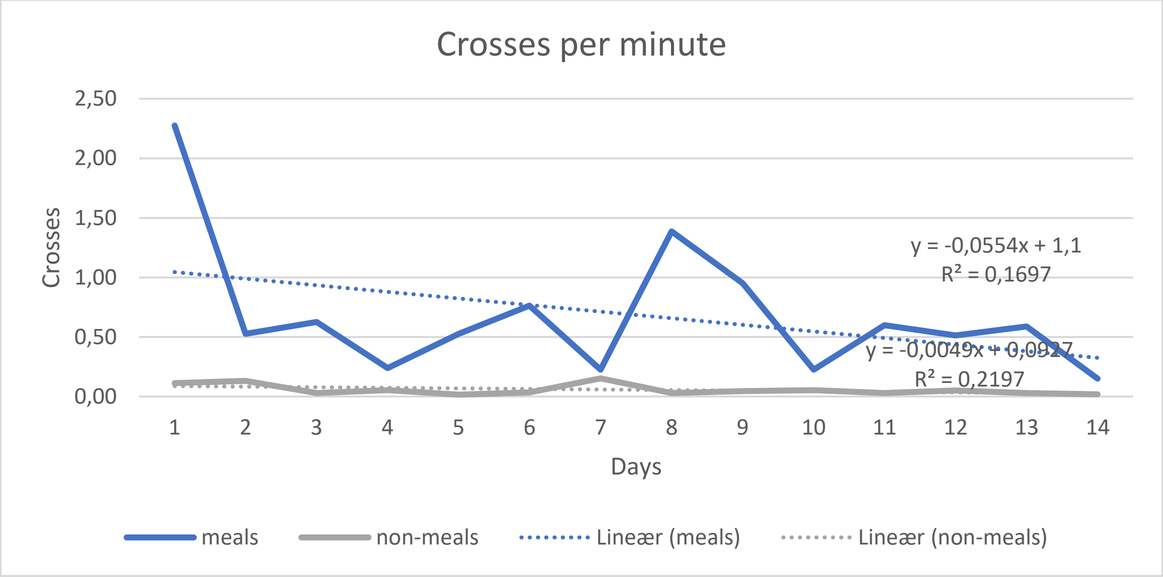


Figure 8: Average crosses per minute during meal and non-meal periods throughout experiment 1.1.

Number of crosses through the hatches was higher during meals than during the long series both in week 1 and 2 (table 4). In week 1 there was an average of 0,74 crosses per minute during meals, and 0,08 crosses per minute during the long series (table 4). In week 2 there was an average of 0,63 crosses per minute during meals, and 0,04 crosses per minute during the long series (table 4).

Table 4: Average crosses per minute during meal and non-meal periods for week on and two in experiment 1.1.

Crosses per minute						
Week #	**Meals**	**Non-meals**	**St.dev. meals**	**St.dev non-meals**	**Var.s meals**	**Var.s non-meals**
1	0,74	0,08	0,70	0,05	0,50	0,003
2	0,63	0,04	0,43	0,01	0,18	0,0002

3.2.2 Exp 1.2 – week 3-4.

3.2.2.1 Total crosses per day

In week 3 and 4, cocurrent through the hatches was tested. After changing the current at day one in the experiment, the total daily crosses decreased from 88 the last day of experiment 1.1, to respectively 0 (after set-out) and 7 on day one and two. The first cross after changing to cocurrent occurred 20 hours after set-out, suggesting the direction of the current though the hatches affects the fish's cross behavior. The total number of crossings increased during the two weeks' period (figure 9). The linear regression analysis gives a slope of 6,1, which means an average increase of 6,1 crosses per day throughout the period (figure 9). The regression analysis also suggests that a variation of 77,5 % can be explained by this model. The increase on daily crosses indicates that the fish is learning how to use the hatches regardless of the direction of the current.

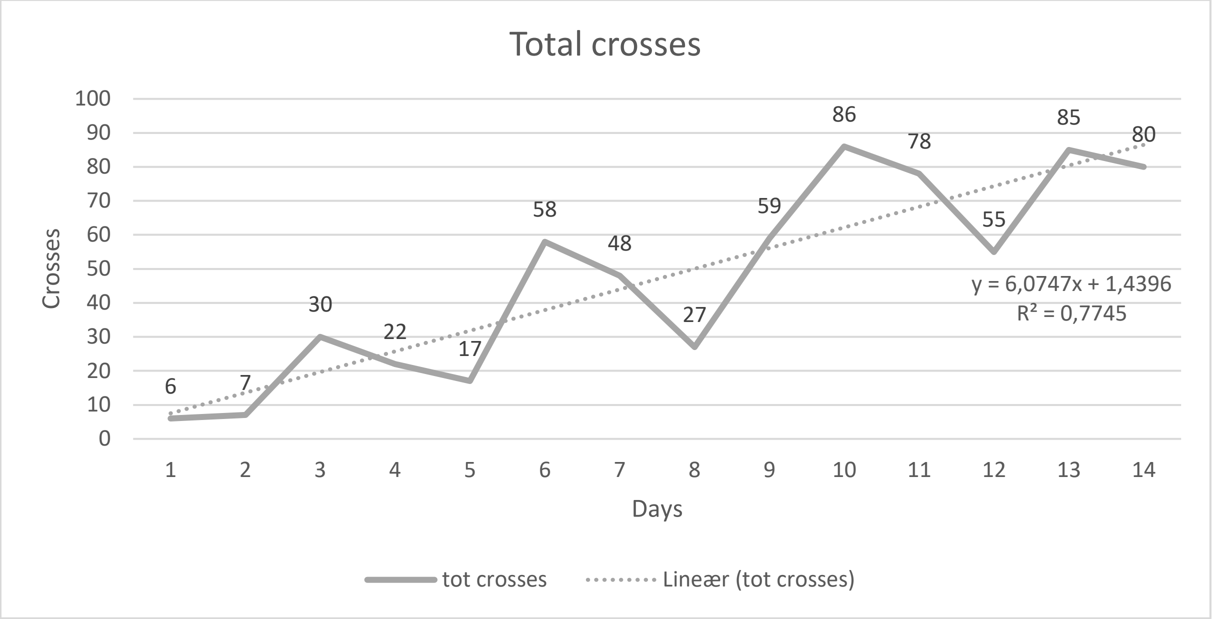


Figure 9: Total crosses per day through both hatches from experiment 1.2 with a regression analysis showing a increase in the total number of crosses per day.

The weekly summary also shows an increase in the average daily crosses over time, with an average of 27 crosses per day the first week and an average of 67 crosses per day the second week (table 5).

Table 5: Weekly summary with average crosses per day, total crosses per week, standard deviation and variance for experiment 1.2.

week #	Average crosses per day	Total crosses	Standard deviation	Variance
3	27	188	20	396
4	67	470	22	464

3.2.2.2 Cross direction through the hatches

Close to 100 % of all crossing were the right way through the hatches, with 99,1 % the first week of the experiment and 100% the second week (table 6). The results show that cocurrent through the hatches does not increase the number of wrong way crosses. Both wrong way crosses appeared during mealtime at day 3 of the experiment, with one cross during the morning meal and the other during the evening meal.

Table 6: Percentage of right-way crosses through both hatches in experiment 1.2.

Week	Right Way	Wrong Way	Total	Right Way Crosses (%)
3 (day 1-7)	217	2	219	99,1
4 (day 7-14)	470	0	470	100

3.2.2.3 Crosses per minute during meal and non-meal periods

The positive slopes of the linear regression trendlines for the periods is in consistent with the increase of total crosses over time (figure 9), where the total crosses during meals shows the greatest increase (figure 10). The results show an increase in cross activity during meals, further the positive trendline in crosses per meal indicates that the number of crosses during meals is the greatest contribution to the increase in total number of crosses over time.

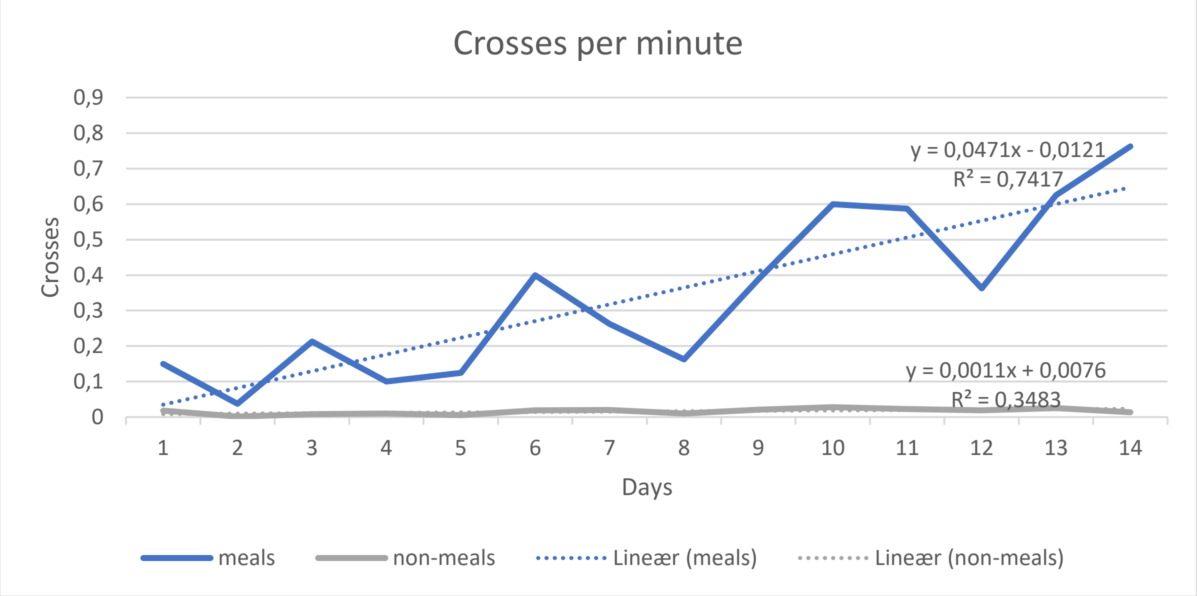


Figure 10: Average crosses per minute during meal and non-meal periods throughout experiment 1.2.

Number of crosses through the hatches was higher during meals than during the long series both in week 3 and 4 (table 7). In week 3 there was an average of 0,17 passings per minute during meals, and 0,01 passings per minute during the long series (table 7). In week 4 there was an average of 0,47 during meals, and 0,02 during the long series (table 7).

Table 7: Average crosses per minute per week during meal and non-meal periods in experiment 1.2.

Crosses per minute

Week #	Meals	Non-Meals	St.dev meals	St.dev non-meals	Var.s meals	Var.s non-meals
3	0,2	0,01	0,12	0,007	0,01	0,00005
4	0,5	0,02	0,20	0,006	0,04	0,00004

3.2.3 Experiment 1.3 – week 5-6.

3.2.3.1 Total crosses per day

In week 5 and 6, smaller hatches with countercurrent were tested. First cross after occurred 2 hours after set-out. The total number of crosses variated greatly throughout the experiment, after weigh in 06.12.18 an increase in crosses occurred (figure 11). The linear regression analysis did not show great change in the daily number of crosses throughout the experiment. Applying separate linear regression analyses for the two weeks gave a decrease of 3,4 crosses per day with a r-square value of 0,19 for the first week (figure 11). For the second week of the experiment the analysis suggested a decrease of 13,2 crosses per day, where 86,4% of the variance could be explained by the model (figure 11).

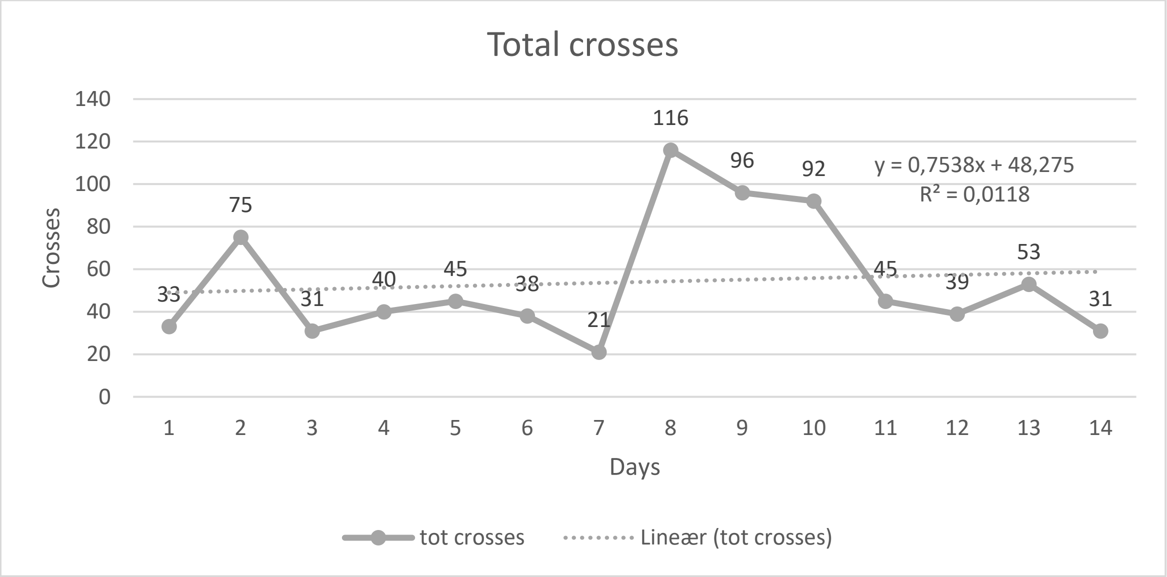


Figure 11: Total crosses per day through both hatches from experiment 1.3.

The weekly summary shows that the total number of crosses increased from 283 to 494, from the first to the second week of the experiment (table 8). There is also a great increase in the variance from the first to the second week, partly explained by the high number of crosses the first hours after set-out and the decrease in the total number of crosses over time.

Table 8: Total crosses, weekly summery.

Week #	Average crosses per day	Total crosses	Standard Deviation	Variance
5	40	283	17	291
6	62	494	35	1205

3.2.3.2 Cross direction through the hatches

Close to 100 % of all crosses were in the right direction through the hatches (from the wider side to the narrow side), with 99,7 % in week 5 and 99,8 % in week 6 (table 9). Meaning close to 100 % of the crosses were countercurrent. The wrong way crosses were registered on day 5 and 10 of experiment 1.3, both during meals.

Table 9: Percentage of right way crosses through both hatches for experiment 1.3.

Week #	Right Way	Wrong Way	Total crosses	Right Way Crosses (%)
5	282	1	283	99,6
6	471	1	472	99,8

3.2.3.3 Crosses per minute during meal and non-meal periods

The slight decrease in the linear regression trendlines for the period (figure 12) is in consistent with the data from total crosses per day (figure 11). The results indicate that the crosses during meals is the greatest contributor to the total number of crosses.

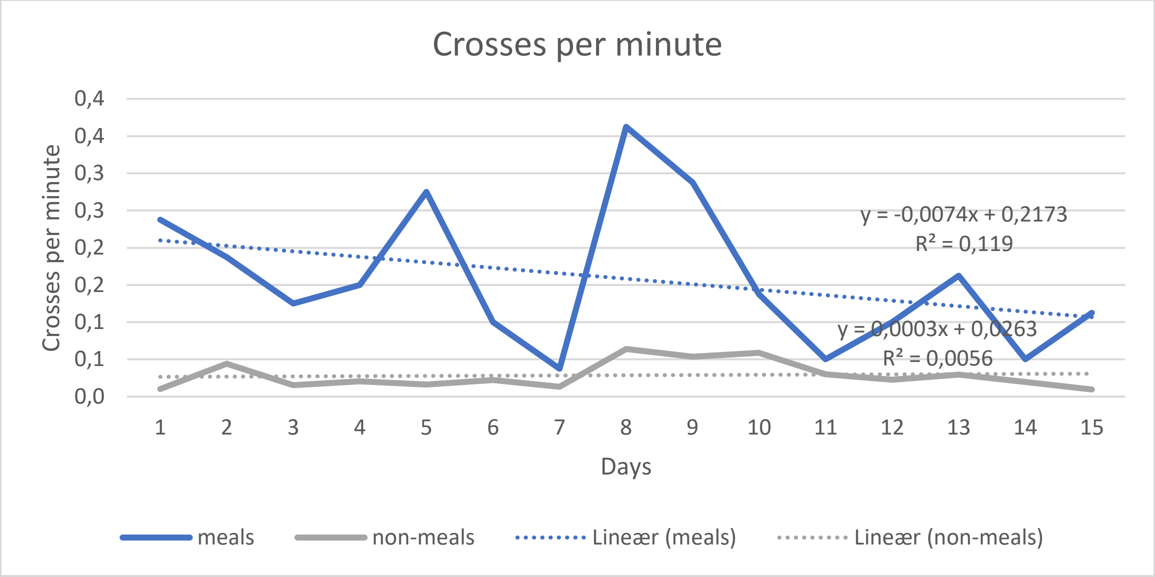


Figure 12: average crosses per minute during meals and non-meal periods throughout experiment 1.3.

Number of crosses through the hatches was higher during meals than during non-meal periods both in week 5 and 6 (table 10). In week 5 there was an average of 0,2 crosses per minute during meals, and 0,02 crosses per minute during the long series, and in week 6 there was an average of 0,2 during meals, and 0,04 during the long series (table 10).

Table 10: Average crosses per minute for meal and non-meal periods per week in experiment 1.3.

Crosses per minute						
Week #	**Meals**	**Non-Meals**	**St.dev meals**	**St.dev non-meals**	**Var.s meals**	**Var.s non-meals**
5	0,2	0,02	0,08	0,01	0,007	0,0001
6	0,2	0,04	0,12	0,02	0,014	0,0003

3.2.4 Total – experiment one

3.2.4.1 Growth Statistics for experiment one

The weight and length for the nine fish increased with time throughout the experiment (table 11). The highest increase in growth was during experiment 1.2, were the average weight increased with 39,4 grams during the two weeks period. In experiment 1.1 and experiment 1.3 the average weight increased with respectively 25,7 and 30,8 grams (table 11). The results indicate that the growth not necessarily is proportionally with the total number of crosses, seeing the total number of crosses were lower during experiment 1.2 and experiment 1.3 than in experiment 1.1 (figure 7, 9 and 10). The coefficient of variation increased from 4,73 % at the start of the experiment to 6,58 % at the end of the experiment (table 11). This indicates a variance in the fish ability to use the hatches and utilize both meals, which results in poorer growth in some individuals. The first week of experiment 1.2, there is a slight decrease in the coefficient of variation compared to the week before (table 11). This indicates a lower variance in the fish's abilities to utilize the new hatch set-up. The first week of the experiment there was a decrease in the average condition factor from 1,3 at set out, to 1,25 at the first weigh in (table 11). Throughout the rest of the experiment there was a slight increase in the condition factor.

Table 11: Growth sampling data from experiment one with average fish weights, lengths and condition factor per week.

01.11.2018	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	231,30	26,39	1,30
Sum	2081,50	237,50	11,34
Coefficient of Variation	4,73	2,28	5,73
Standard Deviation	10,95	0,60	0,07
08.11.2018	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	237,78	26,90	1,25
Sum	2140,00	134,50	11,28
Coefficient of Variation	5,78	2,04	2,70
Standard Deviation	13,75	0,55	0,03
15.11.2018	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	257,00	27,50	1,26
Sum	2313,00	192,50	11,33
Coefficient of Variation	4,78	2,35	3,58
Standard Deviation	12,29	0,65	0,05
22.11.2018	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	276,67	27,89	1,28
Sum	2490,00	251,00	11,48
Coefficient of Variation	5,57	2,50	3,06
Standard Deviation	15,42	0,70	0,04
29.11.2018	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	296,44	28,44	1,29
Sum	2668,00	256,00	11,59
Coefficient of Variation	5,32	2,55	3,52
Standard Deviation	15,76	0,73	0,05
06.12.2018	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	309,67	28,94	1,28
Sum	2787,00	260,50	11,49
Coefficient of Variation	5,67	2,51	3,47
Standard Deviation	17,55	0,73	0,04
13.12.2018	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	327,22	29,33	1,30
Sum	2945,00	264,00	11,66
Coefficient of Variation	6,58	2,56	3,65
Standard Deviation	21,53	0,75	0,05

A linear regression analyses produced a trendline with a slope of 2,4, meaning an average growth of 2,4 grams per day of the experiment (figure 13). The r-square value of the trendline is 0.99, meaning 99 % of the variation can be explained by the model (figure 13).

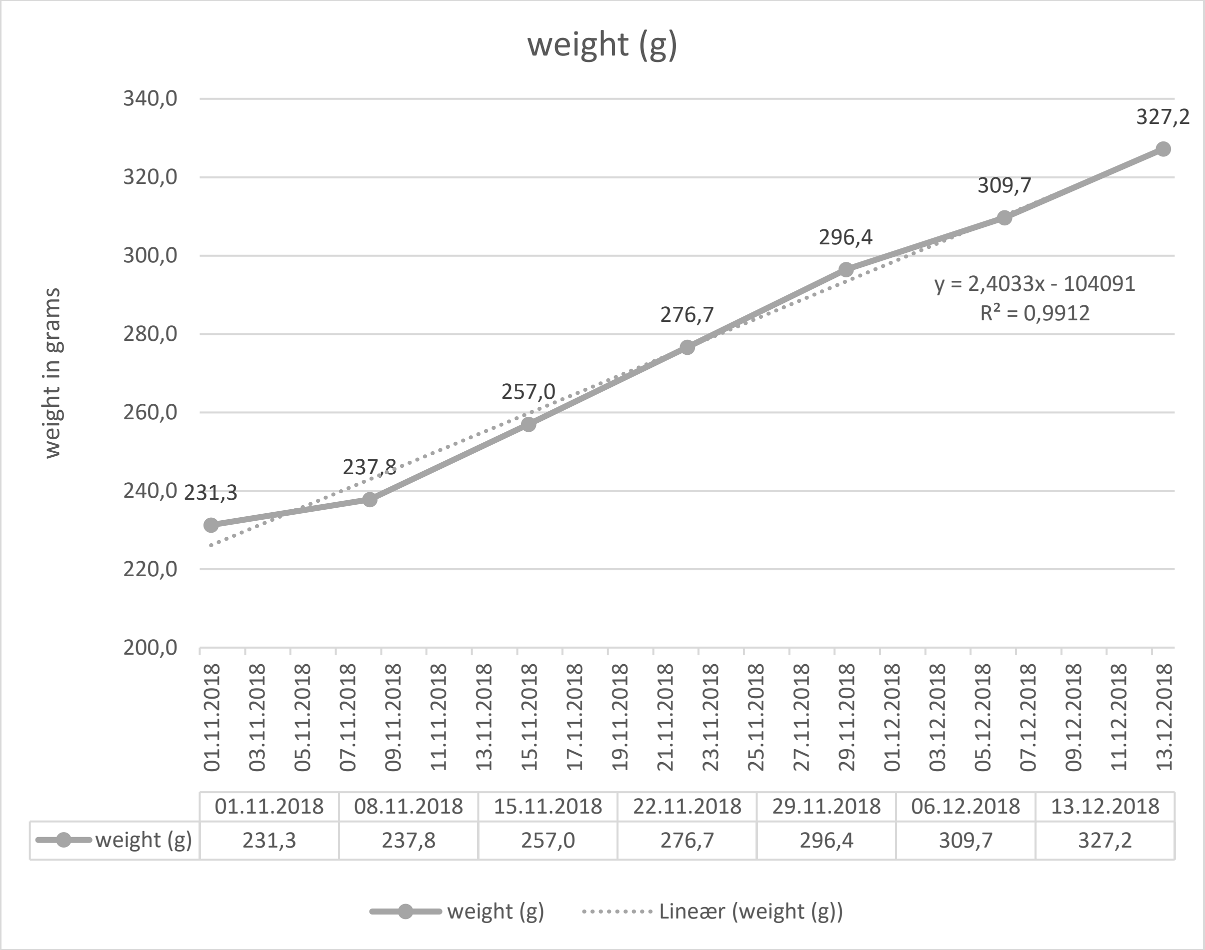


Figure 13: The average fish weight from the weekly weight sampling for experiment one.

The growth sampling date was used to calculate the SGR and FCR for each week during the experiment. The SGR was lower than the expected SGR, and the FCR higher than expected throughout the whole experiment (table 12). This was anticipated for the first period of the experiment, as the fish had to learn how to use the hatches to access both meals. Further in the experiment the SGR was still lower, and the FCR higher than expected, which suggest that not all fish were getting access to both meals (table 12).

Table 12: Expected and calculated SGR and FCR for experiment one.

Date	Expected SGR	Calculated SGR	Expected FCR	Calculated FCR
01.11 - 08.11	1,78	0,40	1	4,68
08.11 - 15.11	1,78	1,11	1	1,63
15.11 - 22.11	1,68	1,05	1	1,62
22.11 - 29.11	1,68	0,99	1	1,73
29.11 - 06.12	1,68	0,62	1	2,77
06.12 - 13.12	1,59	0,79	1	2,19

3.2.4.2 Total crosses per day

The total crosses per day were used to calculate the average number of crosses per day in each week of the experiment. Apart from experiment 1.1, the data shows a decrease in daily number of crosses when changing the tank set-up (table 13). The average daily number of crosses increase in the second week of both experiment 1.2 and experiment 1.3 (table 13). These results are as assumed, as the fish had to learn how to use the new set-up. The results also indicate that the fish preferred using the bigger hatches with countercurrent, seen in the greater number of total crosses during experiment 1.1.

Table 13: Weekly average crosses per day and total crosses per week for experiment one.

Week #	Average crosses per day	Total crosses	Standard deviation	Variance
1	162,14	1133	100,66	10132,14
2	101,00	932	37,71	1422,33
3	26,86	188	19,90	396,14
4	67,14	470	21,54	463,81
5	40,43	283	17,05	290,62
6	61,75	494	34,71	1204,50

3.2.4.3 Cross direction through the hatches

Throughout the experiment, close to 100 % of the crosses were in the right direction through the hatches, from the wider to the narrow side (table 14). Cocurrent through the hatches, or the smaller hatch size, had close to no effect on the share of crossings in the right or wrong direction. Throughout the whole experiment all registered wrong way crosses appeared during meals. This might indicate that the stimuli from the feeder and the available feed in the tank influences the fish's ability to detect the narrow side of the hatch. The wrong way crosses might also be a result of the overall increased activity during meals, resulting in random wrong way crosses.

Table 14: Percentage of right way crosses though both hatches for experiment one.

Week #	Right way	Wrong way	Total	Right way passing`s (%)
1	969	2	971	99,8
2	874	3	877	99,7
3	217	2	219	99,1
4	470	0	470	100,0
5	282	1	283	99,6
6	471	1	472	99,8

3.2.4.4 Crosses per minute during meal and non-meal periods.

Throughout the experiment the average number of crosses per minute were higher during the meals than during the non-meal periods (table 15). In addition to the analysis of crosses during meal and non-meals, a analyze were done to see if there was any increase in activity and number of crossings in conjunction with the meals. The analyze did not show any increase in activity in the last two hours before the meals.

Table 15: Weekly average crosses per minute during meal and non-meal periods.

Week #	Meals	Non-meals	St.dev. meals	St.dev none meals	Var.s meals	Var.s none meals
1	0,74	0,08	0,70	0,05	0,50	0,0030
2	0,63	0,04	0,43	0,01	0,18	0,0002
3	0,18	0,01	0,12	0,01	0,01	0,0001
4	0,50	0,02	0,20	0,01	0,04	0,0000
5	0,16	0,02	0,08	0,01	0,01	0,0001
6	0,16	0,04	0,12	0,02	0,01	0,0003

3.2.4.5 Behavior observations, experiment one

When observing the fish, the focus where on the fish’s ability to cross through the hatches and how the hatch design affected the fish. Typically, the wish would swim along the plexiglass wall searching for the hatch, eventually finding the opening and swim through. The search area variated in depth and length, but the fish would typically start from the bottom and working its way up to the height of the hatch. The fish would not cross from over the hatch, the cross typically occurred from the sides of the hatch or from the bottom of the hatch. When crossing the fish would follow the edge of the hatch, eventually crossing out on the narrow side, or it would cross straight through, not following the edge. There was also observed crosses where the fish would swim in the area nearby the hatch opening, most often on the sides or under the hatch, and after some time cross straight through the hatch. At last, a more direct cross was also observed, where the fish would swim from a greater distance and cross directly through the hatch. In all the crosses described above, there where observations of joint crosses, where a fish would cross though, and a second fish would cross through right after.

Using the cone-formed hatches, the fish would hit the exit (the narrow side) of the hatch pointing into the water column, when searching for the hatch opening. Typically, the fish would change its direction or search at another depth. There were no observations of the fish following the edge of the hatch that was pointing out in the water column, and as a result finding the exit opening and cross though the hatch in the wrong direction. The observed wrong way crosses seemed more random, were the fish would swim nearby the exit opening and randomly getting its nose on the inside of the exit opening and therefore cross through in the wrong direction. When the fish was not crossing, they were typically observed along the side of the tank and the plexiglass divider, often under the exit side of the hatches.

When changing to cocurrent through the hatches in experiment 1.2 the first cross after set-out occurred after a longer period compared to experiment 1.1 and experiment 1.3. In addition, most of the observed crosses where searching crosses and there was a decrease in observations of crosses that were more directly through the hatches. After changing to smaller hatches in experiment 1.3 a new behavior was observed, where the fish would swim into the hatch, turn halfway into the hatch, and swim back out. This behavior was observed one or two times a day throughout the experiment.

3.3 Growth - 13.12.2018 – 17.01.2019

During the 5 weeks period between experiment one and two, the fish where kept in the tank, following the same feeding regimen as in experiment one. The first 4 weeks the fish were kept in the same tank set-up as in experiment one. During this period the average fish weight increased from 327,7 ± 21,5 grams to 393,44 ± 42,48 grams (table 16). The increase in the coefficient of variation for the average fish weight from 6,6 % to 10,3 % indicates a variation in the fish ability to cross through the hatches, meaning some fish's not being able to access both meals.

From 08.01.2019 to 17.01.2019 the new tank set-up was installed, and the fish were moved in and out of the tank two times during that period due to problems with the installation.

Table 16: Growth data with average weights, lengths and condition factor from 13.12.2018 to 17.01.2019.

13.12.2018	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	327,2	29,3	1,30
Sum	2945,0	264,0	11,66
Coefficient of Variation	6,6	2,6	3,65
Standard Deviation	21,5	0,8	0,05
08.01.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	393,44	31,39	1,27
Sum	3541,00	282,50	11,42
Coefficient of Variation	10,80	3,63	4,39
Standard Deviation	42,48	1,14	0,06
17.01.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	387,00	31,44	1,24
Sum	3483,00	283,00	11,18
Coefficient of Variation	10,30	3,85	3,99
Standard Deviation	39,87	1,21	0,05

From the growth data the SGR and FCR for the period was calculated. The SGR for the first period was lower than the expected SGR, and during the last period the SGR was negative (table 17).

Table 17: Expected and calculated SGR and FCR for the growth period.

Date	Expected SGR	Calculated SGR	Expected FCR	Calculated FCR
13.12 - 08.01	1,59	0,71	1	2,31
08.01 - 17.01	1,52	-0,24	1	-6,80

3.4 Experiment 2 – Feeding in a restricted area

3.4.1 Total crosses per day

Average daily crosses during the whole period are lower than in experiment 1. The total number of crosses through the hatches for the whole period was 671, which makes 335,5 crosses in and out of the feeding area, in through hatch 1 and out of the area through hatch 2 (figure 14). Due to some complications when installing the tank set-up, there are no crossing data for the two first days of the experiment (17.01.2019 – 18.01.2019). The trendline from the linear regression analysis suggest a slope of -1,4, meaning an average decrease of 1,4 crosses per day (figure 14). The r-square value is 0,12, meaning that only 12 % of the variation can be explained by the model. The last two weeks of the experiment the fish were kept in the tank with no weigh in to see if no handling would give better growth. On day 13 and 14, were day 13 was the day the weight sampling originally was supposed to happen, there was an increase in the daily number of crosses.

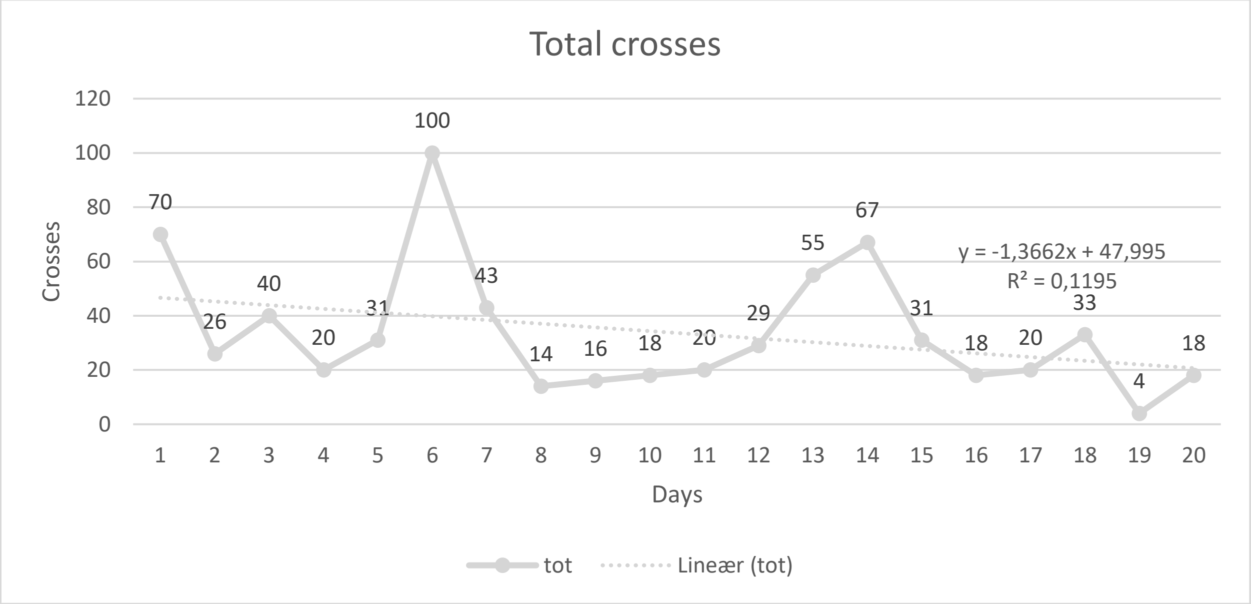


Figure 14: Total crosses per day both in and out of the feeding area for experiment two.

The average crosses per day slightly decreased throughout the experiment (table 18). The standard deviation increased in week 2, because of the increase of daily crosses on day 6 of the experiment.

Table 18: Average crosses per day and total crosses for experiment two.

Week #	Average crosses per day	Total crosses	Standard Deviation	Variance
1	37,4	187	19,64	385,80
2	34,3	240	30,65	939,57
3	32,6	228	21,90	479,62

3.4.2 Cross direction through the hatches

Close to 100 % of all crosses were in the right direction through the hatches, from the wider to the narrow side (table 19). In week two there was two crossings the wrong direction through the hatches. These two crosses appeared on day 7 of the experiment, both during the non-meal period through hatch 2, which is the exit of the feeding area.

Table 19: Percentage of right way crosses, exp.2.

Week #	Right Way	Wrong Way	Total crosses	Right Way Crosses (%)
1	187	0	187	100
2	238	2	240	99,2
3	228	0	228	100

3.4.3 Crosses per minute during meal and non-meal periods

During the first week of the experiment the number of crosses per minute was greater during meals, than during the non-meal periods (figure 15). In week two and three the crosses during meals decreased, except from day 13, 14 and 18 where the crosses per minute was greater during meals than non-meals. Compared to the previous experiment the difference in crosses per minute during meals and non-meal periods is much lower, meaning the crosses appeared more randomly throughout the day. In addition, a analyze were done to see if there was any increase in activity and number of crossings in conjunction with the meals. The analyze did not show any increase in activity in the last two hours before the meals.

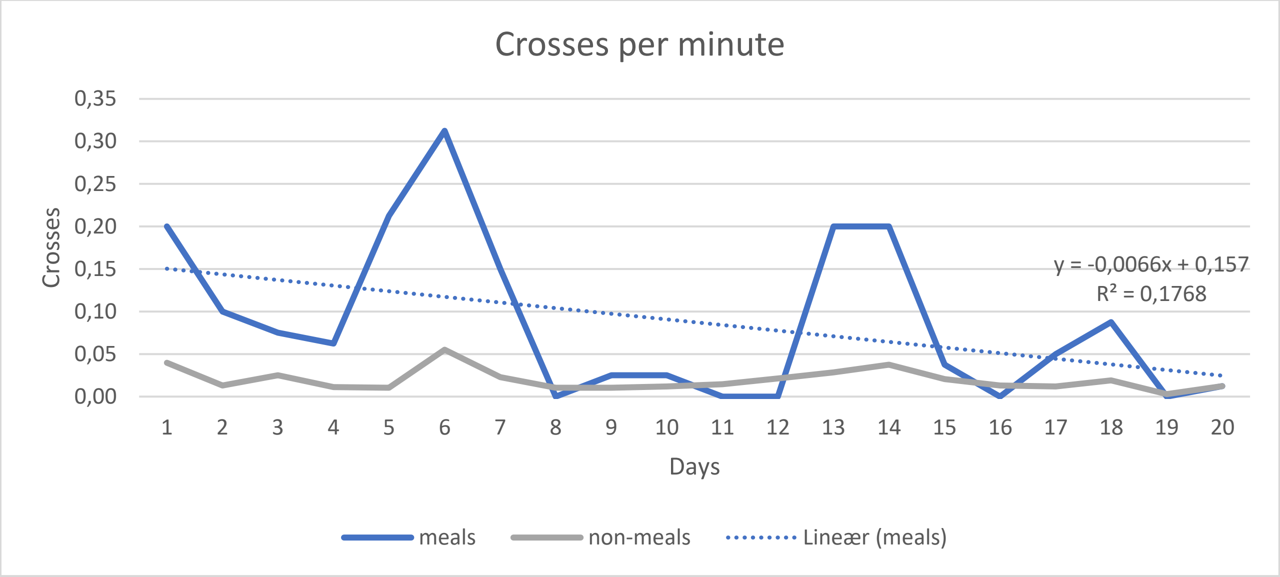


Figure 15: Daily average crosses per minute during meals and non-meal periods for experiment two.

3.4.4 Growth

During the experiment period the average fish weight decreased from 387,0±39,87 grams at the experiment start to 379,44±42,55 grams in the end of the experiment (table 20). Further, the coefficient of variation increased during the same period (table 20), indicating a variance in the fish's ability to access the feeding area and the meals.

Table 20: Growth data with average weights, lengths and condition factor for experiment two.

17.01.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	387,00	31,44	1,24
Sum	3483,00	283,00	11,18
Coefficient of Variation	10,30	3,85	3,99
Standard Deviation	39,87	1,21	0,05
24.01.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	378,89	31,22	1,24
Sum	3410,00	281,00	11,19
Coefficient of Variation	10,31	4,17	5,27
Standard Deviation	39,06	1,30	0,07
07.02.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	379,44	31,83	1,17
Sum	3415,00	286,50	10,56
Coefficient of Variation	11,21	3,85	5,83
Standard Deviation	42,55	1,22	0,07

The growth sampling date was used to calculate the SGR and FCR for each week during the experiment. The SGR was lower than the expected SGR, and the FCR higher than expected throughout the whole experiment (table 21).

Table 21: Expected and Calculated SGR and FCR for experiment two.

Date	**Expected SGR**	**Calculated SGR**	**Expected FCR**	**Calculated FCR**
17.01 - 24.01	1,52	-0,30	1	-5,31
24.01 - 07.02	1,52	0,01	1	151,92

3.4.5 Behavior observations

The cross behavior observed in experiment two was in many ways different than in experiment one. When the fish were not crossing, they were typically located on one half of the tank, known as side one in experiment one. They were located along the sides of the tank, and along the plexiglass wall on the exit side of the feeding area. When crossing, the fish would typically search along the wall on the entrance-side of the feeding area, and cross through the hatch from the sides or from under the hatch. There was also observed a more direct type of cross where the fish would swim in the area near the entrance of the feeding area, and cross direct through the hatch. Joint crosses were also observed. After entering the feeding area, the fish would often stay for several hours. When crossing out of the feeding area, the fish would search along the wall and cross from the sides or under the hatch, the same way as when entering. The observed wrong way crosses did both occur through hatch 2 and were similar to the wrong way crosses observed in experiment one and was perceived as random. When observing more than one fishes in the feeding area at the same time several cases of dominant and aggressive behavior were registered. Typically, the fish staying in the feeding area from start, would chase the second fish entering the feeding area, which often lead to one of them crossing out again. There were also cases of several fish staying in the feeding-area at the same time, with no observation of dominant behavior.

3.5 Experiment 3 – continuously feeding in a feeding area

3.5.1 Total crosses per day

The total daily crosses decreased during the two weeks' period (figure 16). The linear regression analysis gives a slope of minus 1,3, suggesting a slight average decrease of 1,3 crosses per day during the experiment. The number of daily crosses variated from 62 crosses on the second day of the experiment to 7 crosses the last day of the experiment (figure 16). The r-square value is 0,1, suggesting that 10 % of the variation can be explained by the model. Because experiment four were started a few days early, the last week of experiment three only consists of 5 days.

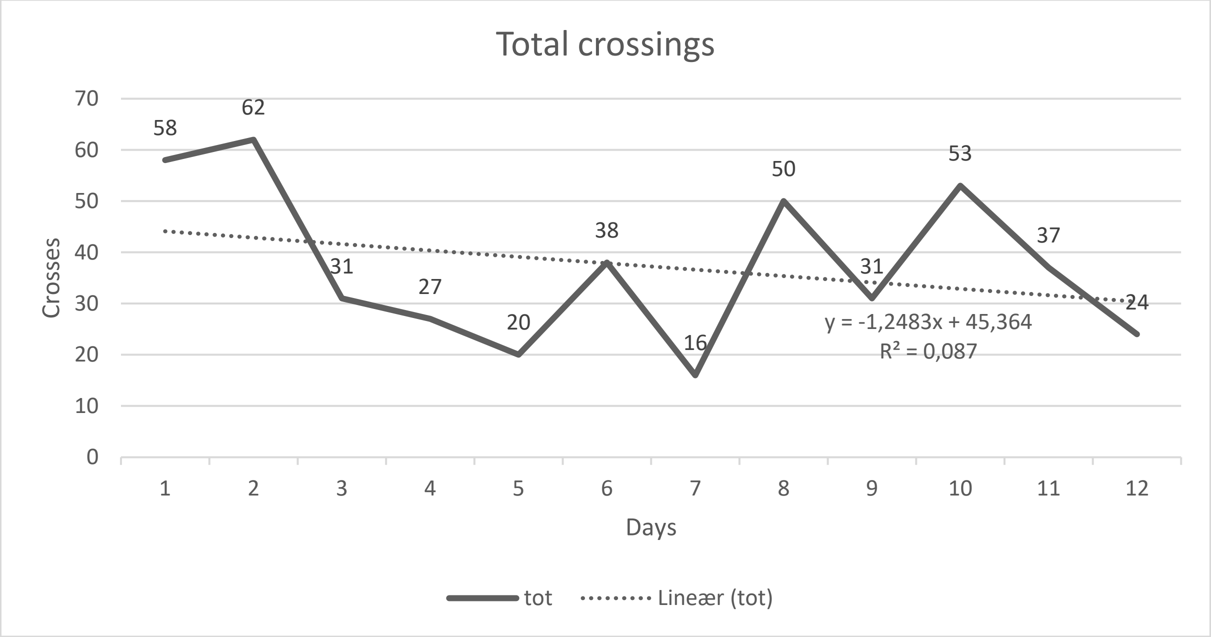


Figure 16: Total crosses per day both in and out of the feeding area for experiment three.

The weekly summary shows a decrease in the average crosses per day, with an average of 36 crosses per day during the first week to 39 crosses per day in the last week (table 22). Due to the last week only being 5 days, the number of crosses is lower in the second week, compared to the first (table 22). Compared to experiment two there was not a significant increase in the number of crosses throughout the period.

Table 22: Average crosses per day and total crosses per week for experiment three.

Week #	Average crosses per day	Total crosses	Standard Deviation	Variance
1 (day 1-7)	36	252	17,9	321
2 (day 8-12)	39	195	12,3	153

3.5.2 Cross direction through the hatches

Close to 100 % of all crosses were in the right direction through the hatches, from the wider side to the narrow side (table 23). Compared to the previous experiments there is a slight increase in the share of wrong way crosses in the second week of the experiment (table 22). All the registered wrong way crosses appeared through hatch 2, the exit-hatch of the feeding area.

Table 23: Percentage of right-way crosses through both hatches for experiment three.

Week #	Right Way	Wrong Way	Total crosses	Right Way Crosses (%)
1	250	2	252	99,21
2	198	4	202	98,02

3.5.2 Growth

The first week of the experiment there was no growth, during the second week there were a slight increase, but mush poorer than expected (table 24). The condition factor was still at an acceptable level (table 24) but was decreasing compared to the previous experiment (table 21). The coefficient of variation calculated for the periods slightly decreased during the experiment (table 24), meaning a slight decrease in the variation of the fish’s ability to cross into the feeding area, getting access to the feed.

Table 24: Growth data with average weights, lengths and condition factor for experiment three.

07.02.2019	Weight (g)	Length (cm)	Condition factor (K)
Average	379,44	31,83	1,17
Sum	3415,00	286,50	10,56
Coefficient of Variation	11,21	3,85	5,83
Standard Deviation	42,55	1,22	0,07
14.02.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	379,44	32,00	1,15
Sum	3415,00	288,00	10,39
Coefficient of Variation	11,39	3,83	5,37
Standard Deviation	43,22	1,22	0,06
19.02.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	383,33	32,00	1,17
Sum	3450,00	288,00	10,50
Coefficient of Variation	10,79	3,58	5,03
Standard Deviation	41,36	1,15	0,06

Due to no growth in the first week of the experiment the SGR and FCR is 0 (table 25). During the last week of the experiment, the calculated SGR was much lower than the expected SGR (table 25). The same appeared for the FCR than was much higher than expected (table 25). These values support the assumptions that there was a variation in the fish’s ability to access the feeding area.

Table 25: Expected and calculated SGR and FCR for experiment three.

Date	Expected SGR	Calculated SGR	Expected FCR	Calculated FCR
07.02 - 14.02	1,52	0	1	0
14.02 - 19.02	1,52	0,0015	1	10,9

3.5.5 Behavior observations

The observations done in experiment three were similar to what was observed in experiment two. The cross behavior occurred the same way as in experiment two, and several cases of dominant behavior was also observed. The wrong way crosses registered in experiment three were all through hatch 2 and was perceived as random. Throughout the experiment there were an increase in the number of observations with more than one individual in the feeding-area at the same time.

3.6 Experiment 4 – feeding in a restricted area with increased flow through the hatches

3.6.1 Total crosses per day

The total number of crosses for the whole four weeks' period was 1400 in and out, making 700 crosses throughout the feeding-area. The number of daily crosses variated greatly throughout the experiment, with a high of 103 crosses on day 15 and a low of 9 crosses on day 5 (figure 17). The linear regression analysis suggests a r-square value of 0,23, meaning that 23 % of this variation could be explained by the model (figure 17). The daily crosses increased during the experiment. The linear regression analysis gives a slope of 1,3, suggesting a daily average increase of 1,3 crosses (figure 17). These results indicate an increase in fish using the feeding area.

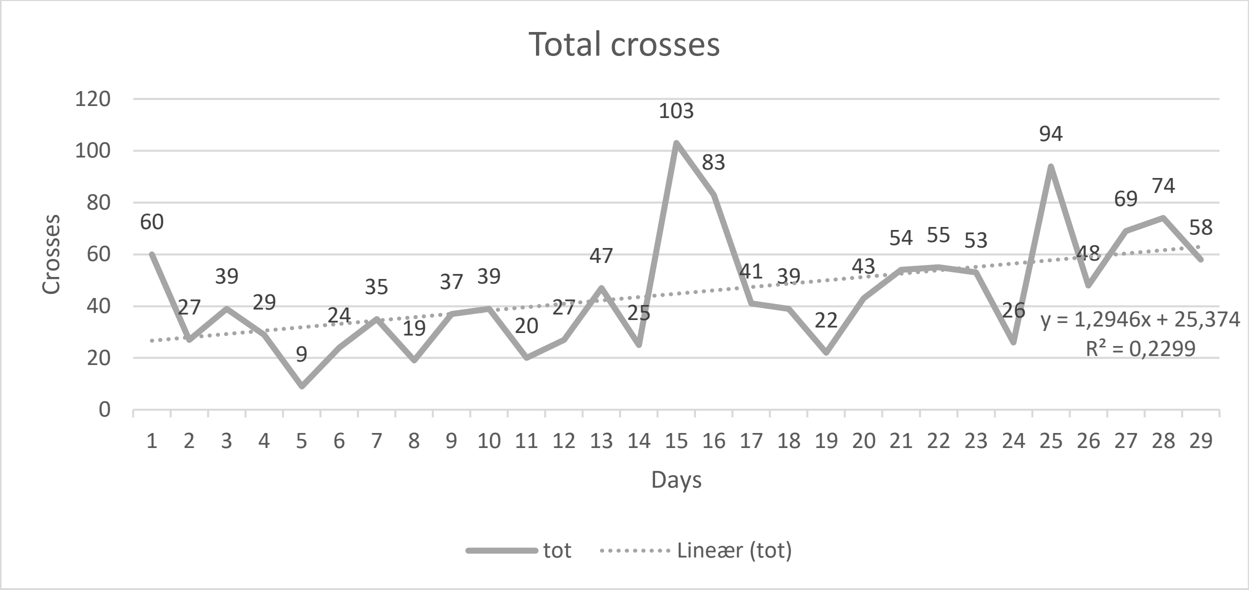


Figure 17: Total daily crosses both in and out of the feeding area throughout experiment four.

The number of daily crosses were used to calculate the average number of daily crosses per week of the experiment (table 27). The average daily crosses increased from 31.86 the first week of the experiment to 59.86 the last week of the experiment (table 27). Compared to the previous experiment there is an increase of the average daily crosses from 39 the last week of experiment three (table 22), to 59,86 the last week of experiment 4 (table 27).

Table 27: Weekly summary with average crosses per day and total crosses per week for experiment four.

Week #	Average crosses per day	Total crosses	Standard Deviation	Variance
1	31,86	223	15,65	244,81
2	30,57	214	10,58	111,95
3	55,00	385	28,21	795,67
4	59,86	419	21,63	467,81

3.6.2 Cross direction

Close to 100% of all crosses were the right way through the hatches, from the wider to the narrow side (table 28). The first wrong way cross in week 2 appeared on day 8 through hatch 1, the next two appeared on day 10 and 11 both through hatch 2. The first five wrong way crosses of week 4, appeared on day 22, all of them through hatch 2. The last two wrong way crosses appeared on day 25, both through hatch 2.

Table 28: Percentage of right way crosses per week for experiment four.

Week #	Right Way	Wrong Way	Total Crosses	Right Way Crosses (%)
1	223	0	223	100
2	211	3	214	98,6
3	385	0	385	100
4	412	7	419	98,3

3.6.2 Growth

The weight and length for the nine fish increased with time throughout the experiment (table 29). The average fish weight increased from 383,33 ± 41,36 grams at the start of the experiment, to 437,89 ± 62,73 grams in the end of the experiment (table 29). In the same period the coefficient of variation for weight increased, indicating an increased variation in the number of fish able to use the feeding area (table 29).

Table 29: Growth data from the weekly weight samplings with average weight, length and condition factor for experiment four.

19.02.2019	Weight (g)	Length (cm)	Condition factor (K)
Average	383,33	32,00	1,17
Sum	3450,00	288,00	10,50
Coefficient of Variation	10,79	3,58	5,03
Standard Deviation	41,36	1,15	0,06
26.02.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	389,44	32,06	1,18
Sum	3505,00	288,50	10,61
Coefficient of Variation	10,40	3,44	4,69
Standard Deviation	40,51	1,10	0,06
05.03.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	395,44	32,33	1,17
Sum	3559,00	291,00	10,51
Coefficient of Variation	10,79	3,79	5,25
Standard Deviation	42,67	1,22	0,06
12.03.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	413,22	32,44	1,21
Sum	3719,00	292,00	10,86
Coefficient of Variation	12,16	3,89	6,07
Standard Deviation	50,24	1,26	0,07
19.03.2019	**Weight (g)**	**Length (cm)**	**Condition factor (K)**
Average	437,89	32,83	1,23
Sum	3941,00	295,50	11,09
Coefficient of Variation	14,33	4,24	7,81
Standard Deviation	62,73	1,39	0,10

The calculated SGR was lower than the expected throughout the whole experiment, due to the variation in the fish ability to access the feeding area (table 30). Although, there was an increase in the calculated SGR over time, meaning there was better growth during the last two weeks of the experiment (table 30). The calculated FCR was higher than the expected FCR throughout the experiment, but with a decrease over time (table 30). These results are indicating an increase in the fish’s ability to access the feed, but there were still individuals not getting access to the sufficient amount of feed.

Table 30: Expected and Calculated SGR and FCR for experiment four.

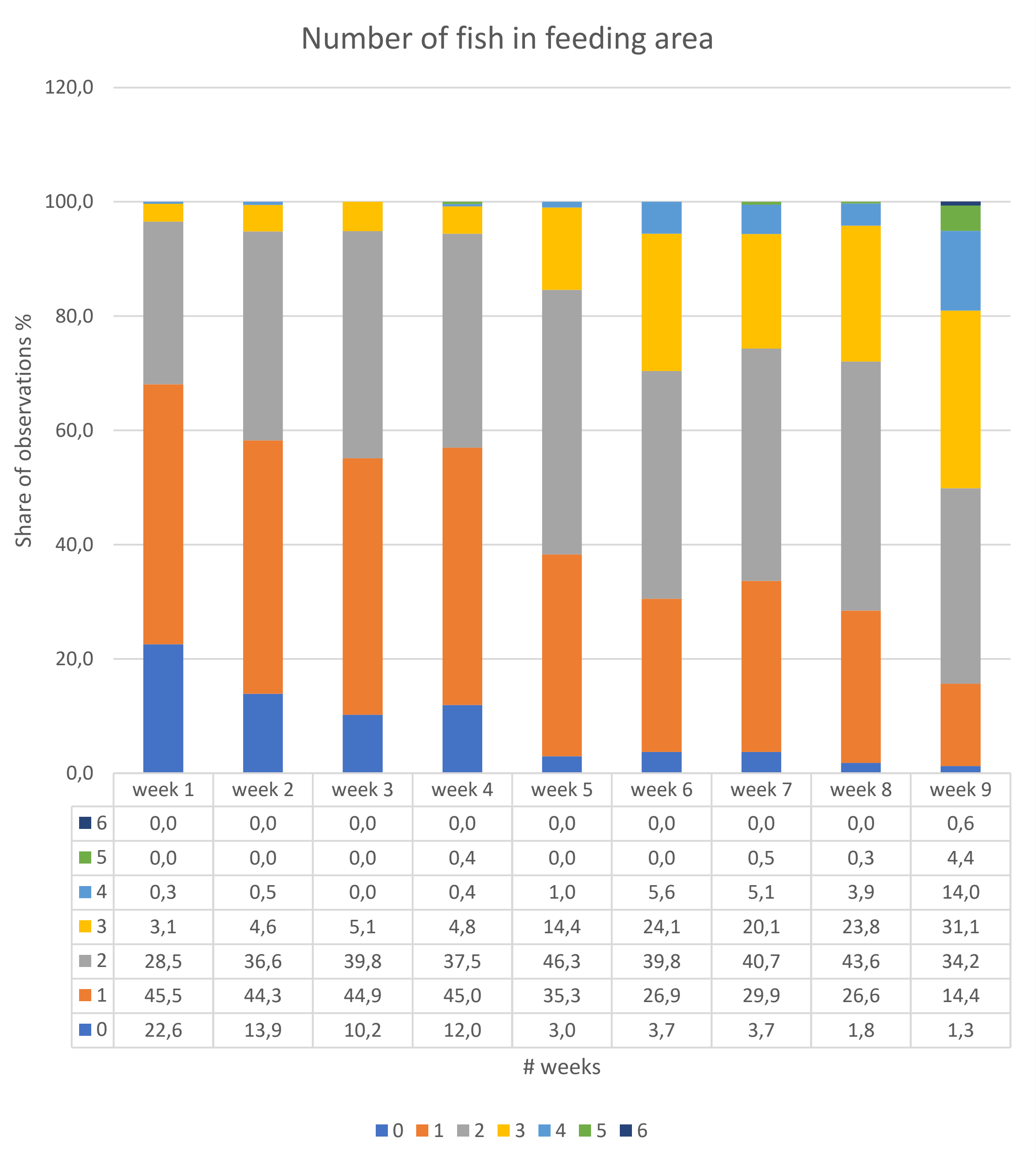
Date	Expected SGR	Calculated SGR	Expected FCR	Calculated FCR
19.02 - 26.02	1,52 %	0,23	1	6,99
26.02 - 05.03	1,52 %	0,22	1	7,23
05.03 - 12.03	1,52 %	0,63	1	2,48
12.03 - 19.03	1,45 %	0,83	1	1,98

3.6.4 Behavior observations

The observations done in experiment four were similar to that observed in experiment two and three, but there was an increase in the occurrence of more direct crosses. After increasing the flow through the hatches, it appeared easier for the fish to detect the them. As in the previous experiments, several cases of dominant behavior were also observed and perceived as random. Throughout the experiment there were an increase in the number of observations with more than one individual in the feeding-area at the same time.

3.7 Feeding area – total exp. 2, 3 and 4

During experiment two, three and four there was an increase in number of fish’s using the feeding area simultaneous. For all crosses, meaning every time a fish crossed in or out of the feeding area, the number of fish’s in the feeding area was counted. These data were used to calculate the share of total observations where it was 0, 1, 2…, or 6 fish staying in the feeding area simultaneous. The share of observations of three and four fish’s in the feeding area increased from respectively 3,1 % to 31,1 % and 0,3 % to 14,0 % from the first to the last week (figure 18). The share of observations of non or one decreased from respectively 22,6 % to 1,3 % and from 45,5 % to 14,4 % from the first to the last week, and in the last week, 4.4 % of the observations was of five fish (figure 18). This result suggests a greater share of the fish being able to access the feeding area and utilize the feed over time.



	week 1	week 2	week 3	week 4	week 5	week 6	week 7	week 8	week 9
6	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,0	0,6
5	0,0	0,0	0,0	0,4	0,0	0,0	0,5	0,3	4,4
4	0,3	0,5	0,0	0,4	1,0	5,6	5,1	3,9	14,0
3	3,1	4,6	5,1	4,8	14,4	24,1	20,1	23,8	31,1
2	28,5	36,6	39,8	37,5	46,3	39,8	40,7	43,6	34,2
1	45,5	44,3	44,9	45,0	35,3	26,9	29,9	26,6	14,4
0	22,6	13,9	10,2	12,0	3,0	3,7	3,7	1,8	1,3

Figure 18: The share of observations calculated from the total number of observations, where 0 to 6 fish were using the feeding area at the same time. 6 out of 9 was the maximum number of fish's observed in the feeding area at the same time.

4. Discussion

4.1 Experiment 1

Close to a 100 % of all crosses through the two hatches occurred the right way from the wider to the narrow side of the hatch. These results are in consistent with the experiments done by (Collins, 1990), showing that the tested hatch-design is suitable for one-way trafficking of fish. To see how different factors would influence the fish behaviour the direction of the current and the size of the hatches were changed. The results demonstrate how the hatch design is of greater importance than the water current to achieve one-way trafficking of fish.

The first cross appeared shortly after set-out of experiment 1.1, showing how the salmon already had learned to cross through hatches to access feed in the previous experiment (Chernin, 2019). The high number of total crosses shows how the hatch design did not affect the cross behaviour of the salmon, in contrast to previous reports has suggested that salmon were afraid of moving through a similar installation; subsurface grading equipment (Fjæra & Skogesal, 1993). Although, there was a slight decrease in the weekly total number of crosses from the previous master thesis experiment done by Chernin (2019), to the first weeks of experiment one, implying that the fish found the new installation more unpleasant to cross. Further, the results show an increase in growth variation and a low, decreasing SGR, and a high, increasing FCR, implying a variation in the fish's ability to utilize the hatches, getting access to both meals.

Changing to cocurrent through the hatches did affect the fish's ability to detect and use the hatch to access the feed. Previous studies of voluntary fish transport where different water flows were tested, the highest flow lead to the best voluntary fish transport (Lekang et al., 1996). Further, the fishes motivation to cross the hatches is dependent on both intrinsic factors, such as hunger and stress, and extrinsic factors such as previous experiences and stimuli from the environment (Fernö et al., 2011). The flow and current through the hatches had several roles, such as carrying the olfactory stimuli from the feed (Sutterlin & Sutterlin, 1971), affecting the motivation to cross, and simply affecting the cross behaviour due to Atlantic salmons favour of swimming against the current (Huntingford et al., 2012). Changing the direction of the current through the hatches limited the stimuli from the feed getting through the hatches and motivate the fish to cross, as demonstrated in the decrease of observations of more direct cross behaviour, from a greater distance from the hatches. However, there was an increase in the number of crosses over time in experiment 1.2, suggesting that hunger is an important motivation to explore the environment (Fernö et al., 2011), and detect the hatches. Throughout the experiments there was a great difference between crosses per minute during meals and non-meal periods, with crosses per minute being much higher during meals. This supports the studies on the flow carrying olfactory stimuli (Sutterlin & Sutterlin, 1971), motivation the fish to cross during the meals. However, crosses per minute was also higher during experiment 1.2 suggesting that getting access to feed simulated the cross behaviour, as a result of previous experience. This is in consistent with previous studies suggesting that salmon exposed to a more complex environment had enhanced foraging performance, as a result of learning from previous experiences (Brown et al., 2003).

The testing of smaller hatches resulted in a decrease in the number of crossings, and behaviour as turning in the middle of the hatch was triggered. These results are in consistent with previous studies showing that voluntary fish transport through gate openings were poorer when decreasing the gate size (Lekang et al., 1996). When increasing the size of the exit of the hatch, it affects the angle and depth from the wider entrance to the narrower exit, making the transition more abrupt. If it is the depth of the hatch, the size of the exit, or a combination that affects the fish behaviour was not solved in this experiment, and further research is needed.

Learning of foraging skills by fish depends on several factors (Warburton, 2003). Intrinsic factors, such as hunger and competition, and extrinsic factors such as previous experiences and stimuli from the environment, affects the fish's motivation to explore their environment (Warburton, 2003). As seen in these experiments with the searching cross behaviour motivated by hunger and competition, and the more direct crosses from greater distance, motivated by previous experiences and stimuli from the flow through the hatches. Further, the number of crosses per minute was much higher during meal than non-meal periods, showing the importance of extrinsic stimuli for motivating the fish to cross. Joint crosses were also observed during the experiment. Learning can also be a result of the fish's ability to utilize information produced by other individuals (Suboski & Templeton, 1989). In social learning the behaviour of one individual attracts the attention of the other individual, often leading to the other individual being exposed to the same stimulus and as a result comes to learn the same behaviour (Brown & Laland, 2003). Social learning can also be a result of imitation where one individual learns by imitating the behaviour of another individual (Brown & Laland, 2003).

Throughout the experiment, several cases of increased activity and number of crosses were observed the first hours after weighing and set out. Studies have shown that anaesthesia with MS-222 induces stress response in Atlantic salmon, leading to high cortisol release rates the first hours after exposure, returning to basal levels after 3 to 4 hours (Zahl et al., 2010). Stress effects induced by handling is also studied, showing an increased cortisol release 3 hours after exposed to handling (Fast et al., 2008). Another study shows how Atlantic salmon smolts who were exposed to stress had increased levels of cortisol concentration within 8 hours, and high plasma glucose levels up to 24 hours after stressor expose (Carey & McCormick, 1998). The induced stress is showed to influence the behaviour and feed intake in Atlantic salmon (Folkedal et al., 2012). The study done by Folkedal et al. (2012), demonstrated a reduced feed intake and food anticipatory behaviour after exposed to stressors. These are reasons to assume that the effects of stress after handling and the anaesthesia have affected the results in these experiments, and in further research should include an acclimation period before collecting data.

In the period between experiment one and two, the fish were held in the tank, with the same set up as in experiment 1.3. During this period the variation in growth increased, amplifying the suggestion of individual variability to utilize the hatches and access both meals.

4.2 Experiment 2 and 3

During the first experiment the hatch were demonstrated to be suitable for one-way trafficking of Atlantic salmon. Further, three experiments were done to investigate the possibilities of getting one fish to enter a feeding area through one hatch, and back into a holding area through another hatch, in one direction only. The hatch design tested in experiment 1.1 were further used in this experiment and installed as an entrance and exit of the feeding area, to imitate the principles of a feeding station. The results show a decrease in the average daily crosses compared to experiment one and the difference in crosses per minute during meals and the non-meal period, decreased throughout experiment 2. Further, there was bad growth with a high, increasing SGR and a low, decreasing FCR throughout the experiment. Also, the cross behavior observed during the experiment would typically be a result of the fish searching along the walls of the feeding area, and more direct crosses from a distance was not observed as often. This implies that the new set up limits the fish’s ability to detect the feeding stimuli from the feeding area. Learning in fish are affected by both intrinsic and extrinsic factors (Warburton, 2003), and limiting the extrinsic stimuli associated with feed effects the fish's motivation to investigate the environment and detect the hatches.

It is known that competition and aggressive behavior is common for Atlantic salmon held in tanks (Turnbull et al., 1998). Previous studies have showed that the aggressive behavior increases at lower population density and smaller groups (Alanärä & Brännäs, 1996). Further, it is demonstrated that competition increases when fish is subjected to defensible food resources, which again leads to unequal food intake among the fish and an increased variation in growth (Ryer & Olla, 1995). A study of the use of self-feeders in rainbow trout, *Oncorhynchus mykiss*, production, demonstrated that use of self-feeders led to a dominance of a few individuals controlling the feeder and therefore affecting the food supply for the whole group (Alanärä, 1996). The same behavior tendency has been observed during the experiments. Several cases of aggressive behavior have been observed, where typically, the fish who is staying in the feeding area chased the next fish entering out of the feeding area. When changing the set up in the tank, making a restricted feeding area, the fish were subjected to more defensible food resources, leading to more aggressive behavior (Ryer & Olla, 1995). This led to a limited ability to access the feed for some fish, and therefore an increased variation in growth throughout the experiment. However, as time passed there were less observation of aggressive behavior, and an increase in the share of observations of more than one individual using the feeding area simultaneous.

When setting up the feeding area, the hatches were placed in different heights to avoid that the fish would swim straight through the feeding-area and to facilitate so the fish would get aware of the feed. During experiment 2 the same feeding regimen as in experiment one were used, with two meals per day. Previous studies have showed that positive reinforcement has a positive effect in the process of learning, where fish has been trained to press a trigger to obtain food (Boujard & Leatherland, 1992). Therefore, continuous feeding was tested in experiment 3 to secure positive reinforcement when the fish crossed into the feeding area. After changing to continuous feeding the results did not show any increase in the number of crosses, nor better growth. There was not any difference in the cross behavior compared to experiment 2. The poor growth, the increase in growth variation and the decrease in the number of daily crosses during the two experiments is suggested to be caused by a combination of limited ability to utilize the feeding station due to the tank set-up limiting the feed stimuli and dominant and aggressive behavior in some fishes.

4.3 Experiment 4

Due to the bad growth results and the increase in growth variation during experiment two and three, the flow through the hatches were suspected to be lower than expected leading to poor stimuli and limiting the fish's ability to detect and utilize the hatches and feeding area. An experiment using food color to get an overview of the current in the tank, revealed close to no flow through the hatches. Therefore, pumps were installed, increasing the countercurrent flow through the hatches. This resulted in an increase in the daily number of crosses compared to the previous experiments. Also, the growth was getting better demonstrated in the increase in the SGR and decrease in FCR. The results support the assumptions that better flow through the hatches would lead the fish to easier detect and utilize the hatches and feeding area. This is in consistent with the discoveries done in experiment one, where changing the direction of the current lead to a decrease in daily crosses. A decrease in aggressive behavior along the fishes over time was observed, which made the feeding area more available.

In further research, its recommended to use individual tagging methods to record the individual growth and feed intake. Due to no tagging the results from the experiments lack the individual monitoring that would make it possible to control which individuals and how many that was using the hatches and utilizing the feed throughout the experiments. To give an idea of how many fishes were accessing the feeding area during the last three experiment, the number of fishes inside the feeding area at each observation was detected and the results were used to calculate the share of observations with several fishes using the feeding area at the same time. From the start of experiment two to the end of experiment four, there was a great increase in the share of observations were several fishes were utilizing the feeding station simultaneous. These results support the assumption of the increased flow making it easier to utilize the feeding area.

5. Conclusion

These experiments have demonstrated that the tested hatches were suitable for one-way traffic of Atlantic salmon. During the first experiment close to all crosses were in one direction only, the right way through the hatch. Further, the shape of the hatch was of greater importance than the water current to get the fish to swim the right way through the hatches. However, countercurrent through the hatches were demonstrated to be preferred by the fish. Throughout experiment one the crosses per minute were significant higher during meals than during the non-meal periods, demonstrating the importance of feed stimuli to motivate and increase the cross behavior. To optimize the hatch size more research is needed. Throughout experiment one the growth variation increased, showing a variation in the fish’s ability to utilize the hatches and access both meals.

Throughout the experiments it was demonstrated that Atlantic salmon was able to learn the behavior of how to cross into a feeding area through hatches to feed, imitating entering a feeding station. The ability to detect and utilize the hatches to access the restricted feeding area, increased flow through the hatches. However, the restricted feeding area were a more defensible food resource which led to an increase in aggressive behavior along the fish. The results show a great variation in the fish’s ability to utilize the hatches, meaning more research is needed, advantageous tagging the fish to enable individual monitoring. The possibilities of limiting the feeding area to a fish at the time, and how to motivate the fish to exit the feeding area when full should be further researched.

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