Brynjar Forseth

Observations of lumpfish at a commercial fish farm during different water current conditions

Masteroppgave i Ocean Resources
Veileder: Kjell Inge Reitan
Medveileder: Anna Solvang Båtnes
Januar 2022

Masteroppgave

NTNU
Norges teknisk-naturvitenskapelige universitet
Fakultet for naturvitenskap
Institutt for biologi

NTNU
Taskforce Salmon Lice

NTNU
Kunnskap for en bedre verden

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Abstract

Salmon lice (*Lepeophteirus salmonis*) is one of the most significant challenges for the Norwegian aquaculture industry and was estimated to account to around 6 billion NOK in 2019. The salmon aquaculture industry uses today cleaner fish as a treatment to control the infestation of salmon lice, and at the northern hemisphere, the cold–water fish Lumpfish (*Cyclopterus lumpus*) has been shown to be a suitable species for Norwegian aquaculture. However, it is limited data of the lumpfish interaction in salmon cages and the performance in different environmental conditions for the lumpfish.

The aim of this study was to examine lumpfish distribution in a full-scale aquaculture farm and determine how water currents affect lumpfish distribution in the cages. In addition, lumpfish was registered in shelter and non-shelter sectors of the cages to evaluate if lumpfish preferred shelter at 3 m and 8 m depth or not. Lumpfish was observed with use of underwater drone over an intensive period from 04th March to 24th April 2022. Dates were chosen based on tidal differences, which creates differences in the current velocity. Environmental parameters were measured either manually (Turbidity) or with sensors (Water temperature, Oxygen, and water current). Statistical analyses were used to test distribution and correlation between lumpfish and water current.

The results showed that lumpfish was significant more abundant at 3m depth than at 8 m depth ($p<0.05$). Lumpfish was also observed more in shelter sectors (28640 observed in shelter sector and 4529 observed in non-shelter sector). Lumpfish at 3 m depth was observed significantly more in the shelter than non-shelter sectors($p<0.05$). Lumpfish observed at 8 m depth was significantly more found in shelter than in non-shelter sectors ($p<0.05$). For spring and neap tide observations at 8 m depth, there were no differences in number of lumpfish in shelter and non-shelter. Lumpfish was not correlated with the different current conditions at neap tide and at spring tide for both depths. Use of lice skirts decreases the water current entering the cages, and this may have contributed to this result and seem to improve the environmental conditions for lumpfish in a commercial aquaculture cage. The result of the present study highlights the complexity of water currents and the distribution of lumpfish in a cage.

Sammendrag

Lakselus (*Lepeophteirus salmonis*) er en av de største utfordringene for norsk havbruksnæring og ble beregnet til å føre med seg kostnader på rundt 6 milliarder NOK i 2019. Lakseoppdrettsnæringen bruker i dag rensefisk som behandling for å kontrollere lakselus, og på den nordlige halvkule har kaldtvannsfisken Rognkjeks (*Cyclopterus lumpus*) vist seg å være en egnet art for norsk havbruk. Det er imidlertid begrensede data om rognkjeksinteraksjonen i laksemerder og ytelsen under ulike miljøforhold for rognkjeksen.

Målet med denne studien var å undersøke rognkjeksdistribusjonen i et fullskala oppdrettsanlegg og finne ut hvordan vannstrøm påvirker rognkjeks distribusjonen i merdene. I tillegg ble rognkjeks observert i rensefisk skjul og ikke rensefisk skjul sektorer i merdene for å vurdere om rognkjeks foretrakk rensefisk skjul på 3 m og 8 m dyp. Rognkjeks ble observert med bruk av undervannsdrone over en periode fra 4. mars til 24. april 2022. Datoer ble valgt basert på tidevannsforskjeller, som skaper forskjeller i strømhastigheten. Miljøparametere ble målt enten manuelt (turbiditet) eller med sensorer (vanntemperatur, oksygen og vannstrøm). Statistiske analyser ble brukt for å teste distribusjon og korrelasjon mellom rognkjeks og vannstrøm.

Resultatene viste at rognkjeks var signifikant mer distribuert på 3 m dyp enn på 8 m dyp ($p<0,05$). Rognkjeks ble også observert mer i Rensefisk skjul sektorer (28640 observert i rensefisk skjul og 4529 observert i soner uten rensefisk skjul). Rognkjeks på 3 m dyp ble observert signifikant mer i Rensefisk skjul enn i sektorer uten skjul. ($p<0,05$). Rognkjeks observert på 8 m dyp var signifikant mer observert i skjul enn i sektorer uten rensefisk skjul ($p<0,05$). For springflo og nippflo på 8 m dyp var det ingen forskjeller i antall rognkjeks i skjul og ikke skjul sektorer. Rognkjeks korrelerte ikke med de forskjellige strømforholdene ved springflo og nippfjære for begge dyp. Bruk av luseskjørt reduserer vannstrømmen som kommer inn i merdene, og dette kan ha bidratt til dette resultatet og synes å bedre miljøforholdene for rognkjeks i en kommersiell merd. Resultatet av denne studien fremhever kompleksiteten til vannstrømmer og fordelingen av rognkjeks i en merd.

Preface

This thesis has been completed at Norwegian University of Science and Technology (NTNU) Trondheim at the department of Biology and been a part of the FOU project Taskforce salmon lice and especially in collaboration with SalMar. The overall objective of the Taskforce salmon project is to establish fundamental knowledge on how the sea lice infest farmed salmon and the mechanisms of how the parasite spread within and between farmed and wild populations of salmonids.

I want to thank my main supervisor at NTNU, Kjell Inge Reitan for helping and supporting me with this master thesis. It has been an honour to discuss and get advice from you. Co-supervisor, Anna Solvang Båtnes, you have been outstanding with your help in the whole proses, from planning the thesis, fieldwork, and all the work with the analysis and finishing this thesis. I could not have done this without your help.

I also want to thank two special persons whit their attribute for this thesis. Kristbjörg Edda Jónsdóttir has helped me to learn a lot of the technology behind measuring water current and teaching me how to use MATLAB. Thank you for sharing the script for the water current. The second person which is an expert in statistic, Lone Sunniva Jevne. Thank you so much for helping with statistically analyses and help to determine which test to use for different data.

My family and friends, thank you for the support. Feedback from persons whit other academic and work backgrounds is helpful. Thank you for the help with the writing process and other useful ways to help and master student.

SalMar has been helping me to find an interesting and relevant study and helping me with all the equipment I needed that I have borrowed from SalMar. I have worked in SalMar since 2018 and have met a lot of great people and learned much about practical aquaculture. SalMar has also provided me with much practical knowledge and advice on what they need to know about lumpfish and how I can transfer knowledge to the company and the industry.

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List of Abbreviations

NMT	Non-Medicine Treatment
ADCP	Aqua Doppler Current profiler
S	Shelter
NS	Non-Shelter
%DO	% Dissolved oxygen in water
PIT tag	Passive Integrated Transponders

1 Introduction

1.1 Norwegian aquaculture

Norwegian aquaculture industry has rapidly grown since its start in the 1960s. it has exported several billion tons of Atlantic salmon (*Salmo salar)* and in 2022 it had an export value of 95 billion NOK (Sjømatrådet, 2023). Norway has a with its long and sheltered coast good conditions for producing Atlantic salmon, including suitable water temperatures (Philis et al., 2018). However, the rapid growth of salmon production in Norway has made many hosts for the parasite salmon lice (*Lepeophteirus salmonis,* Krøyer*, 1837*). Salmon lice are one of the most significant challenges for the Norwegian aquaculture industry and are estimated to cost 6 billion NOK in 2019 (Robertsen et al., 2022, Bergheim, 2012). Norwegian aquaculture is regulated by "*Regulations on combating salmon lice in aquaculture facilities*" (fisheries), 2012) . The regulation aims to reduce the prevalence of salmon lice and the effects on fish in aquaculture facilities and wild salmonids.

1.2 Salmon lice

Salmon Lice is a naturally occurring parasite in the marine environment in the northern hemisphere (Coates et al., 2020). The lice eat the skin, mucus and blood from salmonids which causes wounds, osmotic unbalance, and a higher level of stress (Hamre et al., 2019, Boxaspen, 2006). The lifecycle consists of eight stages (Hamre et al., 2013) that are separated by exoskeleton moult (Sommerset et al., 2021) When the lice find their host, they will attach with frontal filaments, develop to chalimus and start feeding on their host (Hamre, et al., 2019). After the Chalimus stages, the lice will develop into preadults and move around on the fish (Hamre et al., 2019). The last stage of the lifecycle is the adult; at this stage, the adult female develops egg strings (Boxaspen, 2006). According to Norwegian regulations, the limit for the number of lice per fish at a fish farm is set to be 0.5 in adult females and 0.2 in periods during the spring. The different lice limits are set because of the migration of wild salmonids smolts to the ocean (fisheries), 2012).

To reduce the number of salmon lice, it is necessary to use different treatment strategies (Overton et al., 2019). Preventive methods are methods to reduce the rate of new infestations (Barrett et al., 2020). Scientific reports have found that preferred swimming depth for salmon louse copepods (Boxaspen, 2006) and barriers between the louse and the fish in the select water layer would reduce the number of lice inside the cage (Barrett et al., 2020). The most used barrier in Norwegian aquaculture is lice skirts, intending to reduce copepodites attaching to Atlantic salmon (Stien et al., 2012) . Lice skirts are made of either fluid permeable plankton mesh or impermeable membranes (Stien et al., 2012). Lice skirts are attached either inside or outside of the plastic cage (Frank et al., 2015). Lice skirts also come in significant differences in depth and can be from 3 to 12 meters deep (Jónsdóttir et al., 2023). Jónsdóttir et al. (2023) investigated studies of the effect of using lice skirts and the number of lice on different fish farms. Most studies showed promising effects of using lice skirts, but they varied considerably between the fish farms (Jónsdóttir et al., 2023). A snorkel cage is a barrier type closed at the upper water layer to reduce the number of lice attaching to the fish (Wright et al., 2017). Snorkel cages manipulate the salmon to swim deeper and can, with a snorkel which goes up to the surface, refill their swim bladder without risking the attachment of copepods (Barrett et al., 2020, Wright et al., 2017). It is also available in fully closed cages and other technological cages with a semi or closed production unit (Barrett et al., 2020, Nilsen et al., 2020).

Figure 1: Lumpfish eating salmon lice off the Atlantic salmon. Photo: Olivera/Alamy Stock.

When the number of lice on the fish increases, it can be removed with delousing (Nilsson et al., 2023, Overton et al., 2019). Delousing can be done with chemicals, but then more of chemical-resistant lice are increasing (Valenzuela-Muñoz and Gallardo-Escárate, 2016). Fresh water treatment is used to pump fish over to a well boat with freshwater ant kept there for several hours (Overton et al., 2019). Mechanical delousing is removing the fish's lice with pressure flushing, brushing or a combination of these (Gismervik et al., 2017) . The most used method in 2019 was heated seawater. Delousing with heated seawater is done by dipping or bathing the fish in heated seawater before the fish flows back into the cage (Optimar, 2022, Roth, 2016, Overton et al., 2019). In 2019 60 % of the delousing was performed with heated seawater and 26 % with mechanical treatment (Grefsrud et al., 2022, Nilsen et al.). When comparing fish cages with or without delousing, mortality is significantly increased when non-medicine treatment (NMT) is used (Grefsrud et al., 2022, Overton et al., 2019). Using heated seawater is also a discussed method because it is documented that when seawater is heated over 28 °C (Grefsrud et al., 2022), the salmon show signals of pain and perform escape responses (Grefsrud et al., 2022). These methods can significantly challenge welfare with wounds and high-stress levels. It is also observed that higher mortality can last for several weeks after NMT treatment (Gismervik et al., 2017). These methods are some of the reasons for the high mortality in the aquaculture industry. It is also a high welfare issue with an increase of the stress hormone cortisol (Walde et al., 2022). The fish can also risk further infections after a delousing (Grefsrud et al., 2022).

The control method keeps the number of lice under the allowed limit. It is mainly two different methods which are commercially available today. The newest control method uses technological tools such as the lice laser (Stingray, 2022). Lice laser is a new optical delousing, which has limited scientific evidence of efficacy (Coates et al., 2021). One study found the effect of using lice laser to non-significant difference in effect on lice on salmon after 50 days (Bui et al., 2020). The most used control method in Norwegian aquaculture is the use of biological treatment: cleaner fish. Cleaner fish eat lice off the Atlantic salmon and keep the lice numbers down (Figure 1) There are mainly two cleaner fish being used today: Different wrasses (*Labridae)*, wrasses family are temperature sensitive and temperatures under 6°C is not recommended (Sayer and Reader, 1996). Lumpfish (*Cyclopterus lumpus*) are a cold-water fish that prefer temperatures between 3-15°C (Hvas et al., 2018). From 2008 the use of cleaner fish has increased, and in 2019 almost 60 million fish were set out in Norwegian aquaculture sites (Grefsrud et al., 2022). Atlantic salmon has a higher tolerance for water current, and temperature differences than the lumpfish (Imsland and Reynolds, 2022). The differences in the morphology has led to welfare questions of the use of cleaner fish (Stien et al., 2020). In 2018 there was high mortality among cleaner fish, with a total mortality of 42 % in 2018 (Grefsrud et al., 2022).

Also, the lumpfish also has a relatively low aerobic scope compared to the Atlantic salmon and the critical swimming speed for lumpfish is measured to be 1.5 body lengths at 3 °C and 1.67 body length at 9 °C and 15 °C (Hvas et al., 2018). It is reported that 50 % of the fish farms in Norway has a higher current at the aquaculture site than the lumpfish of critical swimming speed (Stien et al., 2020). Norwegian fish farms often use a combination of lice skirts, shelter which are structures inside the net pen that refuges for the cleaner fish to optimize the conditions for lumpfish (Stien et al., 2020). Jónsdóttir et al. (2021) has measured how much the lice skirt and net pens reduce the current and have found a 20 % to 70 % reduction. The variation between sites makes it difficult to quantify the reduction accurately (Jónsdóttir et al., 2023). Much of the variation is due to different site conditions, biomass in the cage and bathymetry for the fish farms (Volent et al., 2020). It is a lack of studies of how environmental parameters affect lumpfish over a longer period and where in the cage the lumpfish are settled. Stenersen (2020) studied the depth distribution between cleaner fish at 1 and 3 m depth and discovered more lumpfish at 3 m depth. Geitung et al. (2020) discovered over an autumn – winter period that lumpfish preferer shallower water from 9 to 1 meter. Further, Overton et al. (2020) recommended future studies to test the performance of lumpfish at different environmental parameters.

1.3 Aim

It is limited date of the lumpfish interaction in salmon cages and the performance in different environmental conditions for the lumpfish (Overton et al., 2020). The aim of this study was to examine lumpfish distribution in a full-scale aquaculture farm and determine how water currents affect lumpfish distribution in the cages. In addition, lumpfish was registered in shelter and non-shelter sectors of the cages to evaluate if lumpfish preferred shelter at 3 m and 8 m depth or not.

This thesis will also observe how different environmental parameters as horizontal water current direction and horizontal water current speed affects lumpfish distribution. We will also observe lumpfish at depths inside and underneath the lice skirt, distribution at different depths and observe where the lumpfish settles in the cage.

2 Materials and Methods

2.1 Study site

The registrations in this thesis were conducted from March to April 2022 at Olausskjæret, a fish farm producing Atlantic Salmon. The farm is in production area 6, placed south in Sulfjorden, southwest of Lyngværet in Frøya (Figure 2). The farm has a production capacity of 3120TN (Barentswatch, 2022). The farm is owned and operated by SalMar Farming AS. The depth distribution at the site varies from 14 to 65 m, with several elevations such as rocks and many deeper areas under the farm. Many islands and islets shelter the farm in all directions. The water current was measured from June to August in 2019 and the main water current was towards northeast with a significant return current towards southwest. The primary water current was classified as strong with a maximum wave height of 2.5 m and water current of 0.98 m/s (Glindø, 2019)

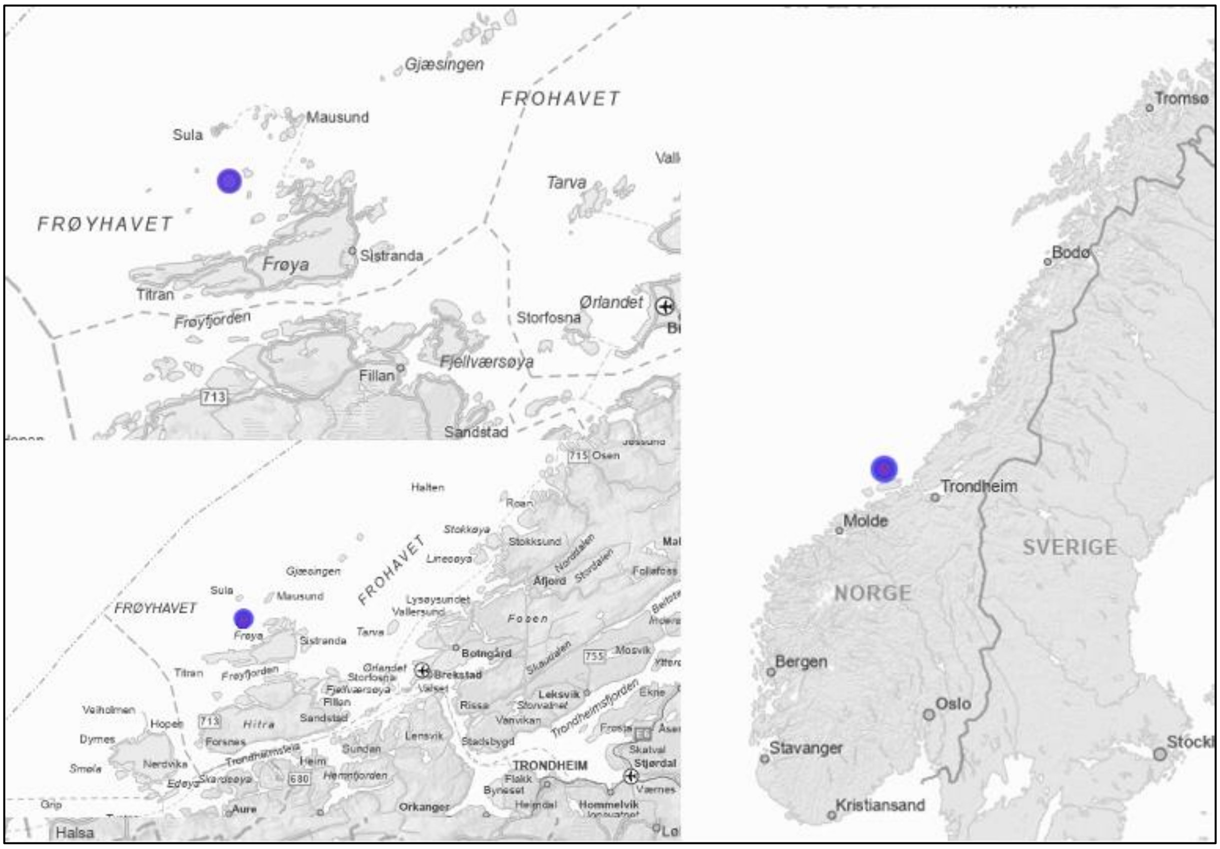

Figure 2: Blue dots show the location of the observation spots at Frøya North, Trøndelag, Norway. The map is modified and originally from Kartverket.

The site has a frame mooring with eight double cages (100x100 m) with five circular floating net cages, 4 cages with fish and 1 cage used as an extra cage for delousing (Figure 3). All the cages had a circumference of 157 m and diameter of 50.9 m. The cages had semi permeable lice skirts 6.5 m deep, with a circumference of 166 m and a mesh size of 340 µm. The nets were cylindrical down to 15 m depth, with a 15 m deep cone (30 m in total). The cages were stocked with Atlantic salmon deployed in April to June 2021, with 3 different smolt groups. Throughout the observation period the farm was managed without delousing treatments and with standard feeding procedures for commercial salmon aquaculture. The observation and counting of lumpfish were performed in the period from March 4 to April 24, 2022.

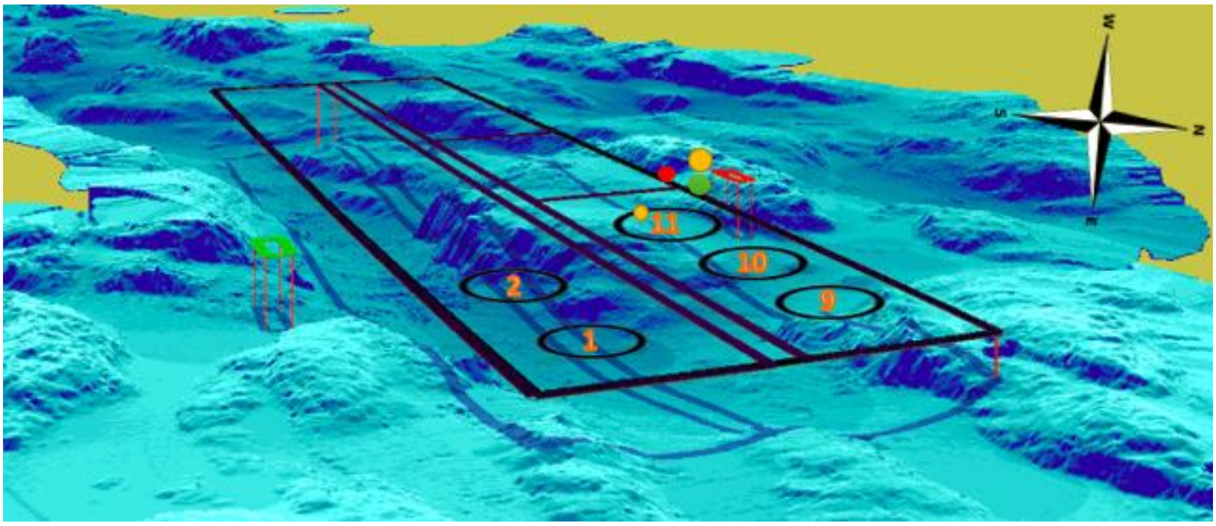

Figure 3: Olausskjæret bathymetry, net cages, and sensors. Cages 1, 2, 10 and 11 contain Atlantic salmon and lumpfish, and cage 9 is used as an extra cage for delousing. Yellow and green dots show the location of the %DO and temperature sensors at 5 and 12 m depth, respectively red cross show the ADCP sensor. Green circle shows the temperature and oxygen sensors at 12 m depth. Map from: OLEX

2.2 Recording of environmental parameters

Water temperature (°C), Water turbidity (Secchi depth; m), Dissolved oxygen (%DO), water current speed (cm/s), and water current direction (°) was measured at the fish farm (Figure 3) (Nortek, 2020). All the different sensors were weekly inspected and cleaned during the observation period. The sensors measuring water temperature and %DO logged with an interval of 10 minutes. The sensors were connected to a receiver which uploaded the data to the cloud. Data were registered at 3 m (Water current, temperature, Dissolved oxygen), 5 m (Water temperature, dissolved oxygen), and 12 m depth (water temperature and dissolved oxygen). Water turbidity was measured using a seawater Secchi disk, a 30 cm disk with black and white quadrants. It was lowered into the sea until it was not observed, and the depth of the disappearance was then the measure of the turbidity of the water. The water current speed and directions were measured by an Aquadopp current profiler (ADCP 600 Khz) produced by Nortek (2020) The ADCP was programmed using software from Nortek, (2022). The program used standardized settings for ADCP to measure downwards. The ADCP was programmed with 10 minutes measurement interval throughout the observation period. The mooring of the instrument is shown in figure 5 and the instrument was deployed in November 2021 and retrieved in May 2022. The instrument was regularly checked for biofouling, vertical movement, and the mooring structure was checked. The instrument was placed 50 m from the feed barge and 50 m from cage 11 (Figure 3). It was placed 2 m under the sea surface and measured every 2 m to the bottom 36 m. The ADCP did not measure the upper 3 m, because the ADCP was placed at 2 m depth and the first 1m measurements were discarded due to low quality. Figures 4 and 5 show images of the mooring which was used during the observation period.

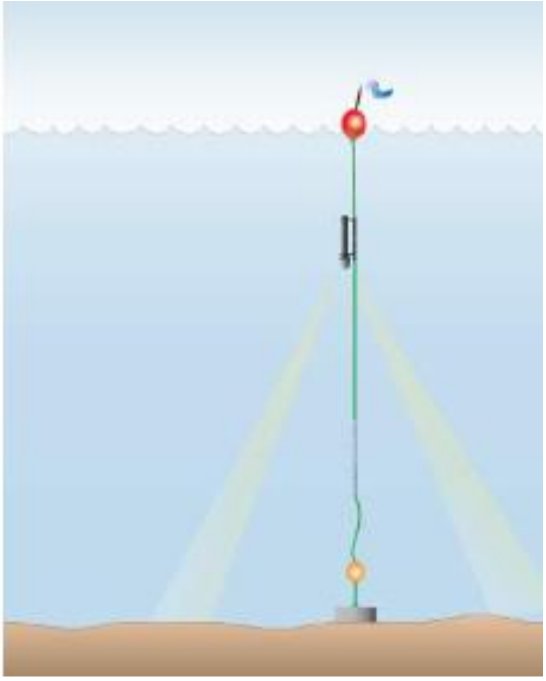

Figure 4: The mooring of the water current profiler (ADCP) The anchor is at 36m depth, the chain is at 30m and ADCP at 2m (Nortek, 2020)

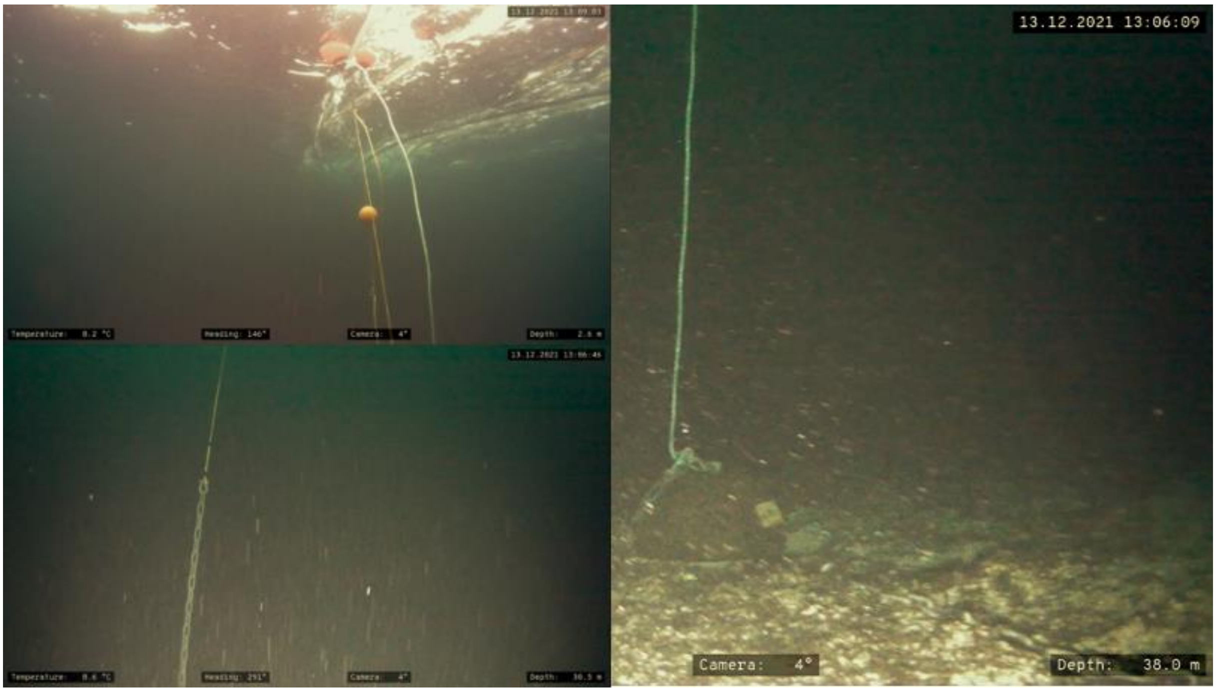

Figure 5: Mooring of the ADCP. Upper left: top of the mooring, Lower left, Between chain and rope. Right left, seafloor and 500 kg weight. Photo: Brynjar Forseth.

2.3 Tidal cycle

The tidal difference at Olausskjæret varied with over 200 cm between maximum high tide and minimum low tide under spring tide for the observation period. Neap tide had a variation of 50 cm between high and low tide. Figure 6 illustrates the tidal cycle in the period and the water level. The data was downloaded from Kartverket (2022) and modified.

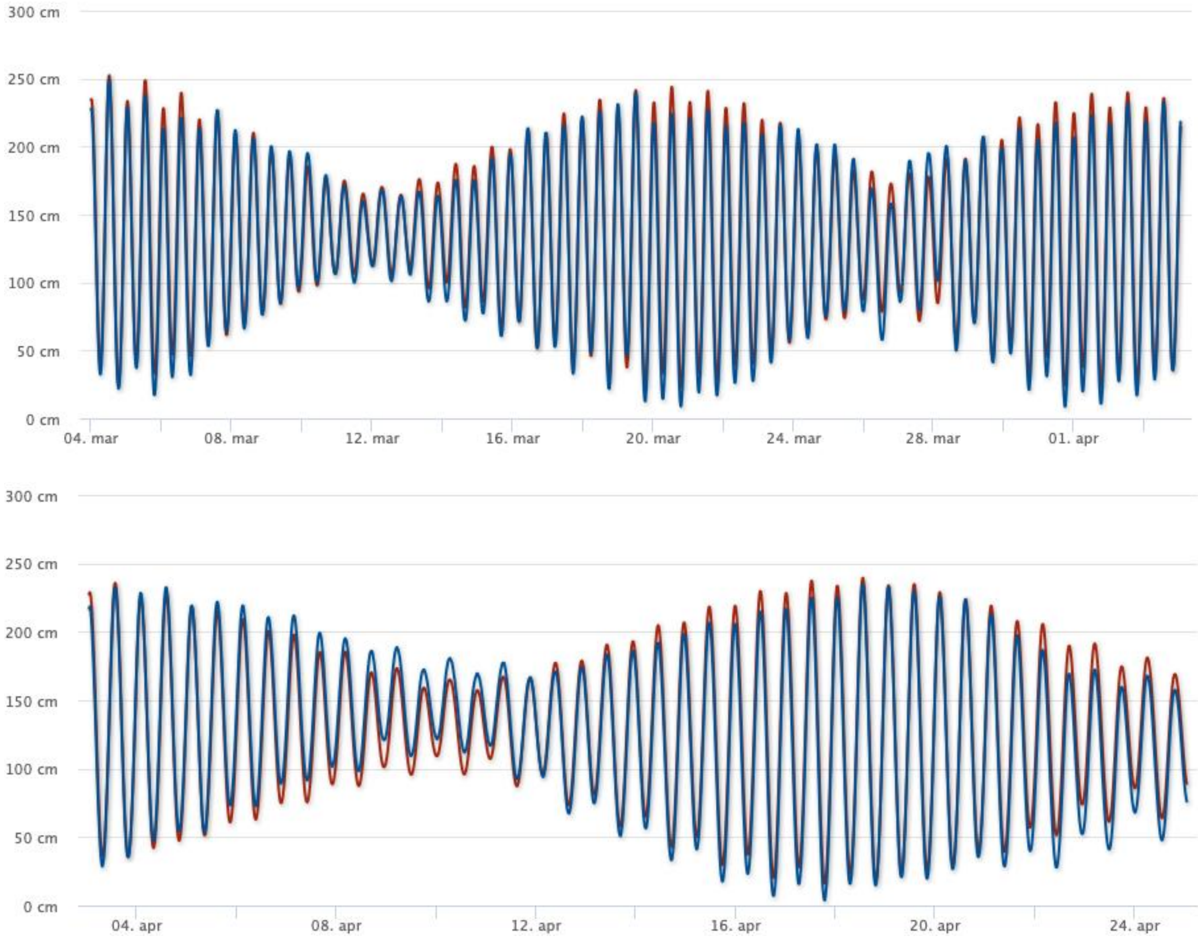

Figure 6: Tidal cycle and water level under the observation period. Illustrational purpose Source: (Kartverket, 2022).

2.4 Observation and registration of lumpfish

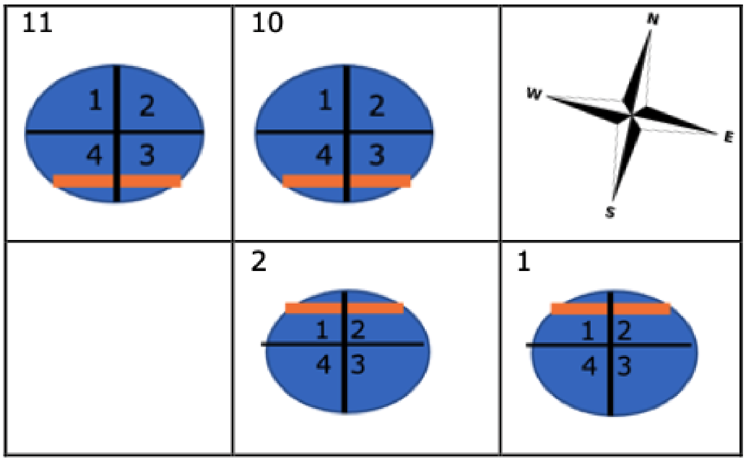

Figure 7: Overview of net cages where lumpfish were observed. Observations were divided into sectors 1-4 in each net cage. Orange lines represent lumpfish shelters

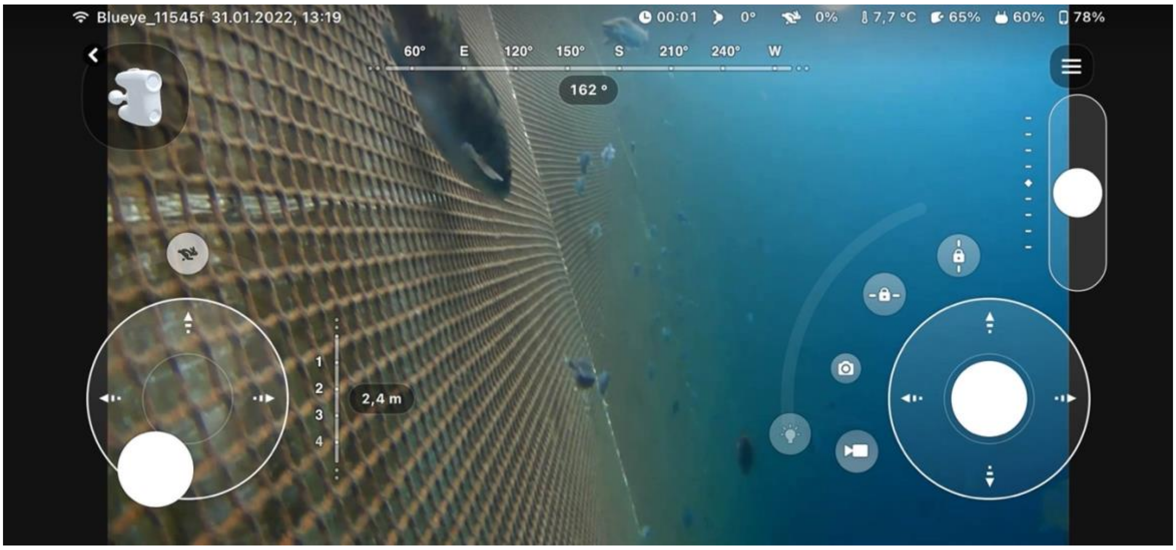

Figure 8: Blueeye under the observations of lumpfish. Photo: Brynjar Forseth

The lumpfish were observed at 3 and 8 m depth alongside the net in 4 cages. Lumpfish was sorted into 4 different sectors after the observation where two of the sectors was at the shelter and two at non-shelter sectors (Figure 7). Lumpfish were observed with a BlueEye Pioneer underwater drone (BluEye, 2022). All the observations with Blueeye were standardized to take 20 minutes, and the drone had 1.5 m distance from the net, and a width sight of 1,5 m. That is, lumpfish were observed between 1.5-4.5 m and 6.5 to 9.5 m depth. To keep the Blueeye stabilized and orientated, a depth stabilizer was used to keep the drone at decided depth Figure 8. The net cages have 60 side ropes (15 per sector) and counting side ropes on the net was done to keep track of the drone position. The registration dates were chosen based on the water tides, with spring and neap tide, observed every week for two months (Figure 6, Table 1). After each observation, the lumpfish in each sector, at 3 and 8 m depth were counted using QuickTime Player at 0.25x speed and a digital number counter. Number of lumpfish

The total number of lumpfish in each cage is shown in Table 1. This estimate is based on the numbers from counters in the well boats and mortality under the period. The lumpfish were set out in December 2021 with an initial mean weight of 40 ± 4,5 g. The site had 4 shelters per cage as shown in Figure 7, of which 2 were in use at a time, and a 14-day rotation for cleaning the shelters. The cleaning process was done by lifting two shelters diagonally, and the biofouling was dried, and the shelter were cleaned. The lumpfish was fed with an automatic feeder placed between the shelters. The lumpfish was fed according to the feed producers' recommendations.

Table 1: Estimate of lumpfish in the different cages. Start number is from the well boat and further based on mortality numbers from the fish farm.

Cage	04.03	12.03	20.03	27.03	02.04	10.04	18.04	24.04
1	6501	4911	3805	3472	3057	2785	2391	2165
2	7431	5234	4118	3763	3239	2958	2556	2306
10	8650	6250	5408	5184	4730	4422	4107	3988
11	6566	4428	3730	3549	3198	2790	2486	2212
Total	29148	20823	17061	15968	14224	12955	11540	10686

2.5 Data treatment and statical analysis

Raw water current data was downloaded from Aquacurrent and then transferred to Seareport, Version 1.1.11, Nortek (2021). The program removed velocity spikes, boat noise and other disruptions for the ADCP. Data not approved in Seareport was removed and not used. After removing error sources and quality checked, it was further investigated in MATLAB (2021),Version r2021a 9.10.0.1602886, to visualize water speed and directions. Data extracted for each observation day was water current speed (Maximum, minimum and mean) and water direction (Maximum, minimum and mean). The direction the current of in each sector was also noted in excel in minutes. The environmental data was collected 4 hours before observation and 4 hours under the observation to sample data which could affect the observations. The 4 different sectors were under the data treatment adjusted to 2 sectors, with and without shelter, to account for differences in shelter placement at the different cages (Figure 7).

The data was systematized using Microsoft Excel (Corporation, 2022), version 16.8) filtrated in a nested design and sorted to smaller parameters of different variables. In addition, a Pivot table was used to make illustrations, to compare and observe similarities and trends between the datasets. The data was then analysed in SigmaPlot (2022) and SPSS (Version 28.0.1.0 142, IBM SPSS Statistic). To compare means between two populations, the T-test was applied. Testing and comparing means for the different populations, the analysis of variance ANOVA was used. Both the T-test and the ANOVA require that the data is normally distributed and do not contain any skewness. To test the data for normal distribution, a Shapiro-wilk normality Felt test was applied. (Shapiro and Wilk, 1965). Majority of the data was not normally distributed, so it was attempted to transform the distribution data to normal. After testing the data (after natural log transformation), it was still not normally distributed, and it was decided to use a non-parametric test (Mann-Whitney U test) to investigate the lumpfish distribution in the cage. We discussed to either use a one or two tailed test because we assumed that the lumpfish would be more distributed in the shelter than in the non-shelter sector. It still was necessary to use the strength in a two tailed test for shelter and non-shelter. Significance level was set to be $p < 0.05$. Lumpfish is shown as % observed lumpfish for 8 and 3 m depth.

For testing the correlation between observed lumpfish and the different current conditions it was also used a non-parametric test with a correlation coefficient (Table 2). It was determined to use a Kendall's Tau test since it would fit the smaller data set and the many tied ranks variation of the data better than the Spearman's correlation rho would do (Xu et al., 2013).

Table 2: Correlation Coefficient, (Xu et al., 2013, Hinkle et al., 1979).

Strength of Correlation	Interpretation
.90 to 1.00 (-.90 to -1.00)	Very high positive (negative) correlation
.70 to .90 (-.70 to -.90)	High positive (negative) correlation
.50 to .70 (-.50 to -.70)	Moderate positive (negative) correlation
.30 to .50 (-.30 to -.50)	Low positive (negative) correlation
.00 to .30 (-.00 to -.30)	Negligible correlation

3 Results

3.1 Environmental parameters

The temperature at different depths (°C), Dissolved oxygen levels (%DO) and water turbidity (m) were registered for all the observation dates (Table 3). The water temperature at the different depth showed less than 0.5 (°C) variations throughout the observation period, and the measurements done at 3 m depth were therefore used for the analyses (Table 3). Water temperature increased from 5.9 to 7°C over the observation period. %DO was different between 5 and 12 m depth with a reduction of the oxygen concentration at 12 m outside of the cage compared to 5 m depth.

Table 3: Environmental parameters at the observation dates. Temperature 3m, dissolved oxygen at 5 m and 12 m depth outside of the cage, dissolved oxygen 5m depth inside of the cage, and turbidity (Secchi depth) outside of the cage.

Date	Temperature 3m (C°)	%DO5m outside of cage	%DO 12m outside of cage	%DO 5m inside cage	Turbidity (m)
04.03.2022	5.9	110	99	106	11
12.03.2022	5.9	108	94	104	11
20.03.2022	5.9	116	95	111	7
27.03.2022	6.1	107	78	101	7
02.04.2022	6.1	116	101	110	10
10.04.2022	6.0	110	100	108	8
18.04.2022	6.5	125	105	110	9
24.04.2022	7.0	118	109	112	6

3.1.1 Horizontal water current direction and speed

The horizontal water current direction (Figure 9), for the observation period, measured by the ADCP showed a mean water current direction of 154°. It was a low variation of the water current direction under neap tide and spring tide. Lowest mean was measure to 79° and highest mean was measured to 197°, measured over a period of 51 days. There were no significant differences observed in current direction between the water layers 0-20 m depth at the different observation dates. The current direction changed more rapidly under 22.5 m depth and the direction over was more stable. It was also compared between 3 and 8 m depth and there were not found a difference in the water current direction.

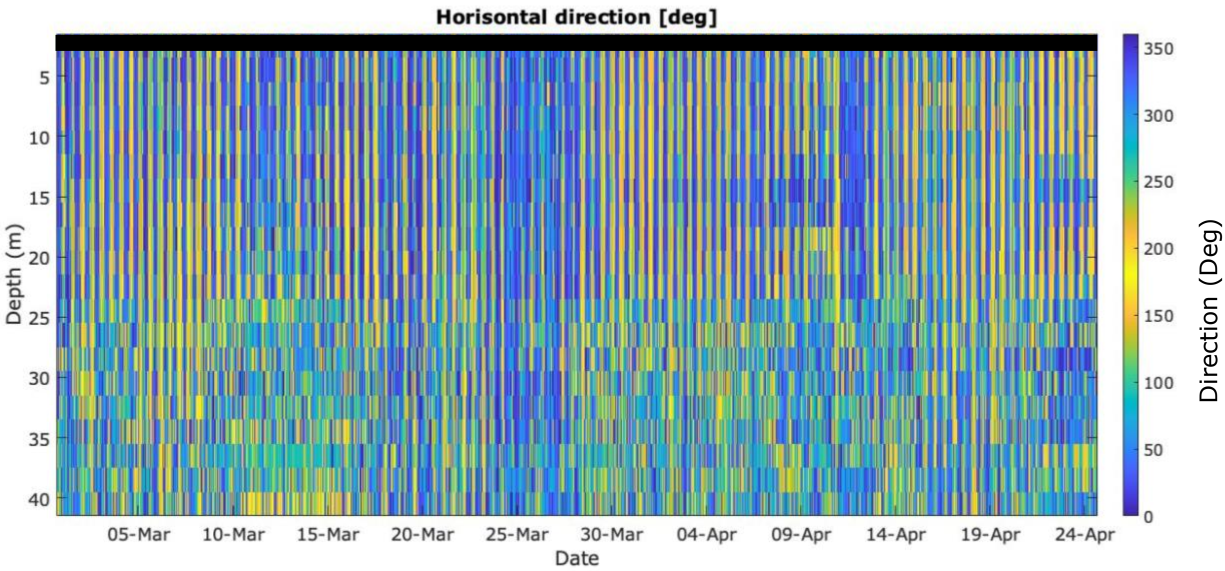

Figure 9: Water current direction(deg) measured by the ADCP in the observation period. X-axis: date, Y-axis: depth (m). Colour bar: degrees. For position of the sensor, see Figure 3.

The horizontal water current speed (Figure 10) was measured under the observation period by the ADCP. Mean water current speed at 3 m depth for the observation period was measured to 12.3 cm/s (Figure 9 & 10). Highest measured current speed at 3 m depth was measured at spring tide 20.03 with 36 cm/s from direction 321°. Lowest water current speed (cm/s) at 3 m depth in the period was measured to 0.1 cm/s from direction 140°, measured under neap tide at 12.03. For 8 m depth the highest measured water current speed was measured under neap tide at 12.03 with a 38 cm/s speed at direction 221°. The mean water current speed for the period at 8m depth was measured to 13.8 cm/s in speed from direction 135°. The lowest water current speed at 8 m depth was measured 12.03 at 0.2 cm/s from 90°.

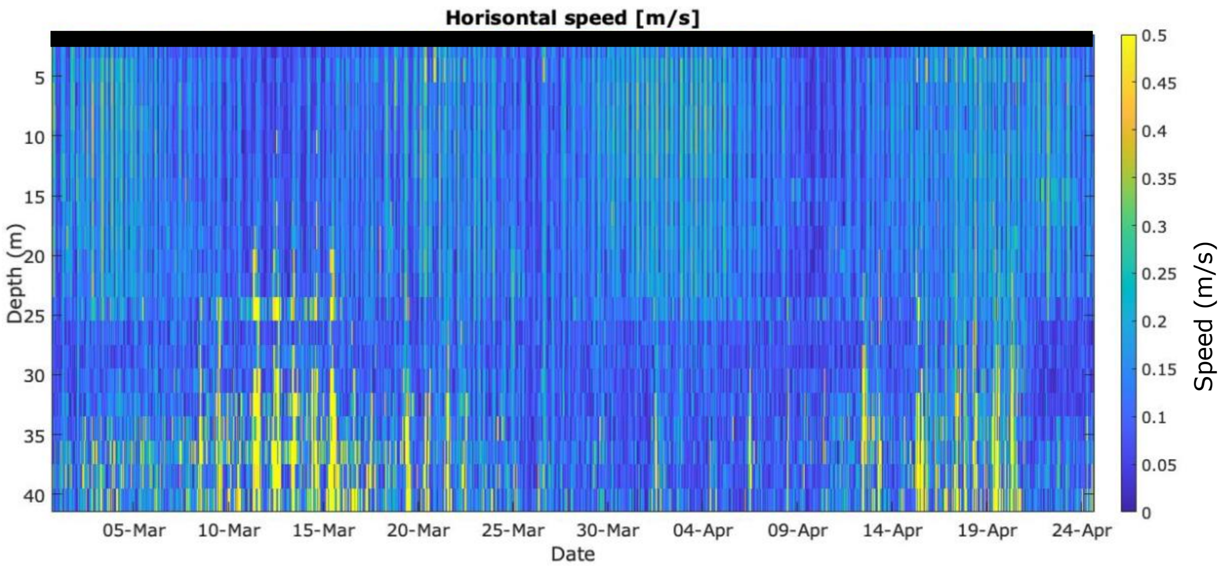

Figure 10: Horizontal current speed (m/s) measured by the ADCP through the observation period. X-axis: date, Y-axis: depth. Colour bar: current speed. For position of the sensor, see Figure 3.

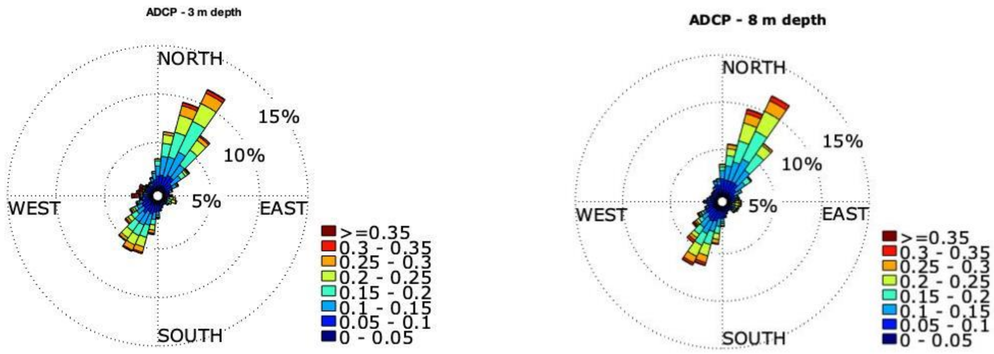

Figure 11: A) Current speed (m/s) and current direction (°) at 3 m depth. B) Current speed (m/s) and current direction (°) at 8 m depth. For the observation period, measured by ADCP. For placement of the ADCP see figure 3.

To visualize the horizontal current speed in the observation period, polar roses for 3 m and 8 m depth (Figure 11) were made. The main water current is from Northeast with a low Southwest current in return for both depths. At 3m a slightly broader current (in degrees) is observed compared to 8 m depth.

3.2 Lumpfish Distribution

The lumpfish distribution was observed in the period 4th March to 24th of April and sorted in shelter and non-shelter (Figure 12). In total, during the entire period, 33 638 lumpfish were observed: 33169 lumpfish at 3 m depth and 469 lumpfish at 8 m depth. There was a significant difference ($p=0.001$) between registered lumpfish at 3 m depth and 8 m depth.

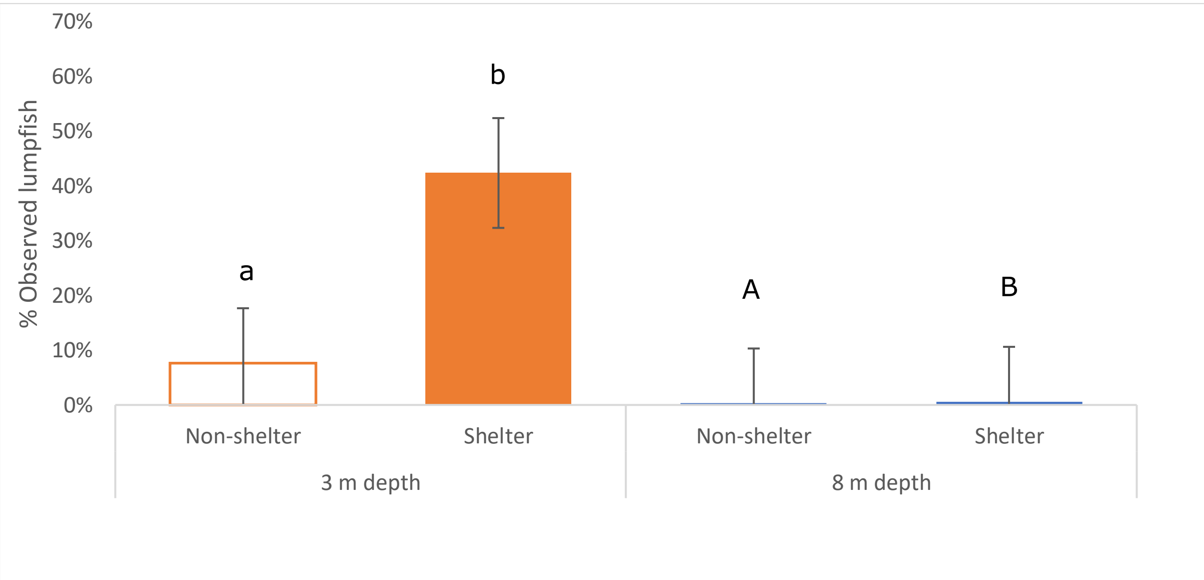

Figure 12: *Distribution of lumpfish in the sectors, at 3 and 8m depth, in % mean of all observed lumpfish (± SE). Significant differences between shelter and non-shelter sectors are marked* by letters. 3 m depth (lowercase letters) and 8 m depth. (Uppercase letters) have not been compared to each other.

The lumpfish distribution at 3 m depth in the period was as follows: 28640 observed lumpfish in shelter sector and 4529 observed lumpfish in non-shelter sector. There was found to be significant difference between shelter and non-shelter sectors (p=0.001), using Mann-Whitney U test. For 8 m depth 371 lumpfish were observed in the shelter sector compared to 98 lumpfish in non-shelter sectors. Significant difference between shelter and non-shelter sectors (p=0.001) were found at 8 m depth (Figure 12).

3.2.1 Lumpfish in sectors at different tidal conditions

During spring tide, in total 17716 lumpfish were observed at 3 m depth: 14929 lumpfish in shelter and 2787 lumpfish in non- shelter sectors. Again, significantly (p=0.001) more lumpfishes were observed in the shelter sectors (Figure 13). At 8 m depth, 75 lumpfish were counted in non-shelter sectors, and 223 lumpfish in shelter sectors, not significantly (p=0.798) found to be more lumpfish at shelter sectors.

During neap tide at 3 m depth, 15453 lumpfish were observed in total: 13711 in shelter and 1742 in non-shelter (Figure 13). A significant (p=0.001) difference between shelter and non-shelter sectors at 3 m depth was found. At 8 m depth 171 lumpfish were counted: 148 in shelter and 23 in non-shelter sectors and found non-significant at 8 m depth was found between shelter and observed lumpfish (0.675).

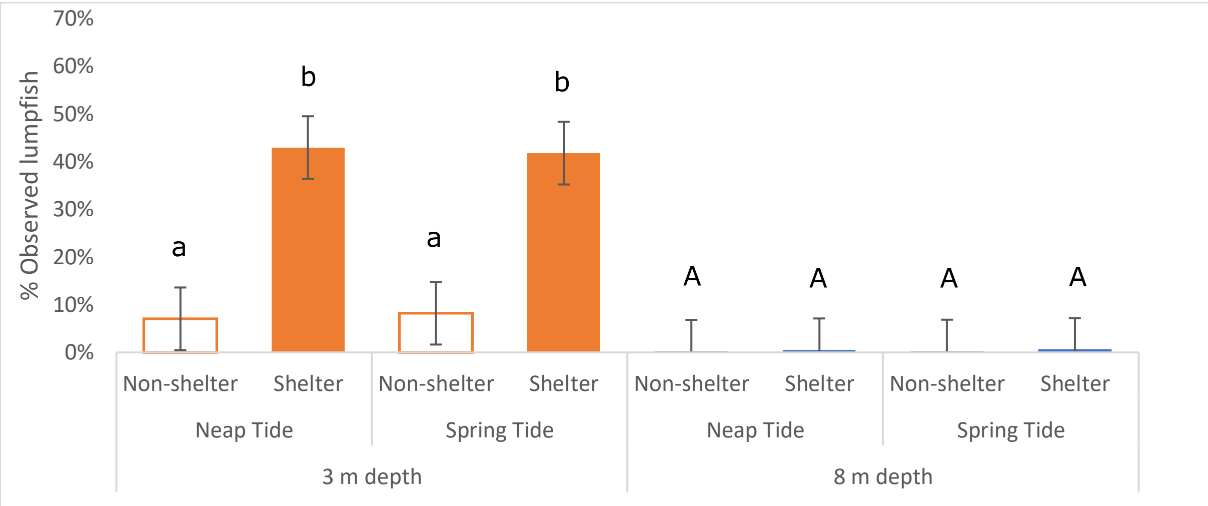

Figure 13: The distribution of lumpfish at 3 and 8m depth within shelter and non-shelter sector divided into Neap tide and Spring tide for the whole observation period as % mean of all observed lumpfish (± SE) at each registration date. Significant differences are indicated by letters. 3 m depth (lowercase letters) and 8 m depth (uppercase letters).

3.3 Tidal conditons affecting lumpfish distribution

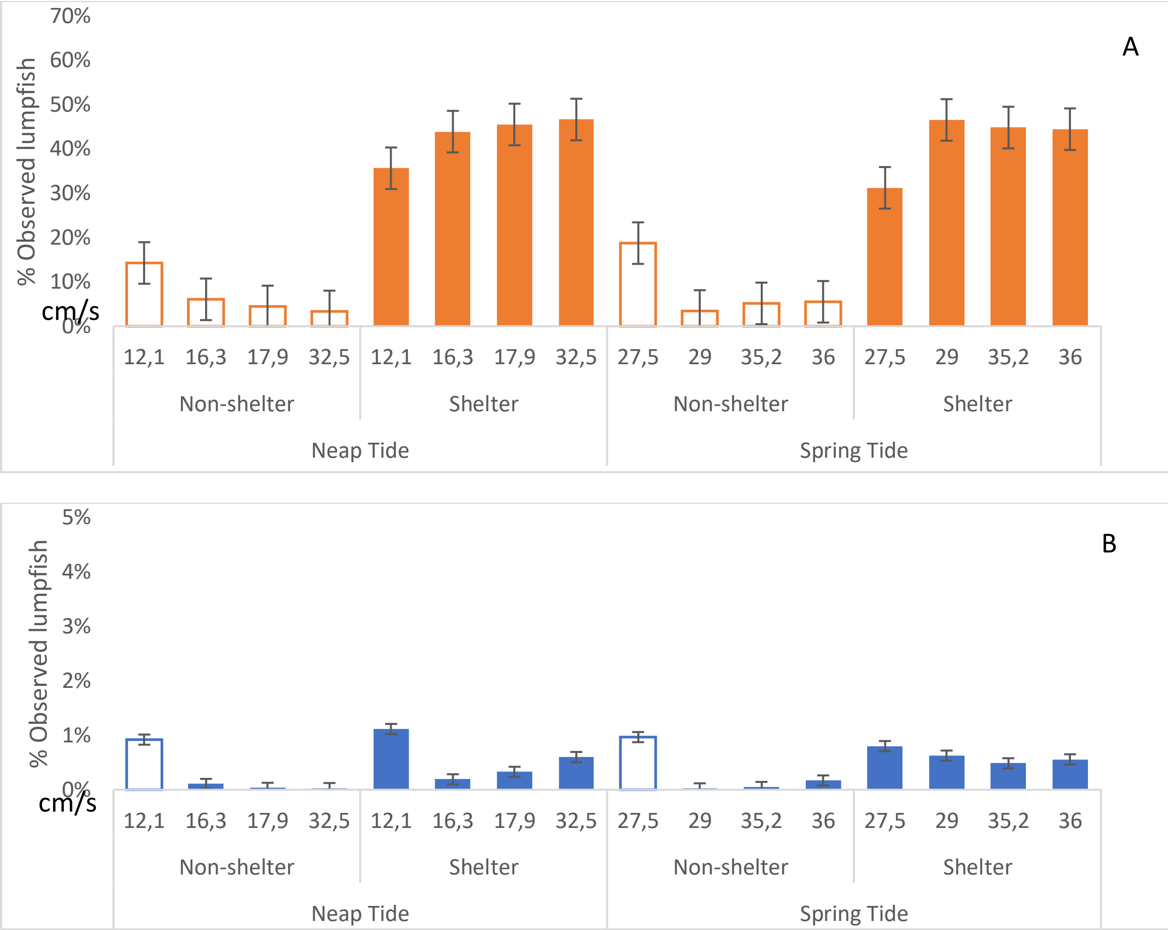

Figure 14: Mean % observed lumpfish (± SE) at A) 3 m and B) 8 m depth, at maximum measured current (indicated on X-axis in cm/s) during the observation period, during neap tide and spring tide and for different sectors. Note: different y axis range in A) and B).

In total, during the entire period there is no correlation between water current speed and observed lumpfish at 3 and 8 m depth (0.02, $p>0.05$) (Figure 14). There is also non correlation between duration (min) the current entered the sector and observed lumpfish (0.098, $p>0.05$) (Figure 15).

During spring tide, the correlation between observed lumpfish at 3 m depth and maximum measured current showed non correlation (0.18, $p>0.853$) (Figure 14). Again, non-correlation with observed lumpfish and current in each minute for different sectors (0.041, $p=0.641$) (Figure 15). At 8 m depth, observed lumpfish and measured current speed, showed negligible correlation (0.252, $p=0.013$). Measured current in each minute for different sectors also showed negligible correlation (0.109, $p=0.247$)

During neap tide, the correlation between observed lumpfish at 3 m depth and maximum measured current showed non correlation (-0.127, $p=0.181$) (Figure 14). Again, negligible correlation with observed lumpfish and duration (min) the current entered the sector (-0.072, $p=0.418$) (Figure 15). At 8 m depth, observed lumpfish and measured current speed were non correlation (-0.127, $p=0.214$). duration (min) the current entered the sector also showed non correlation (0.058, $p=0.550$)

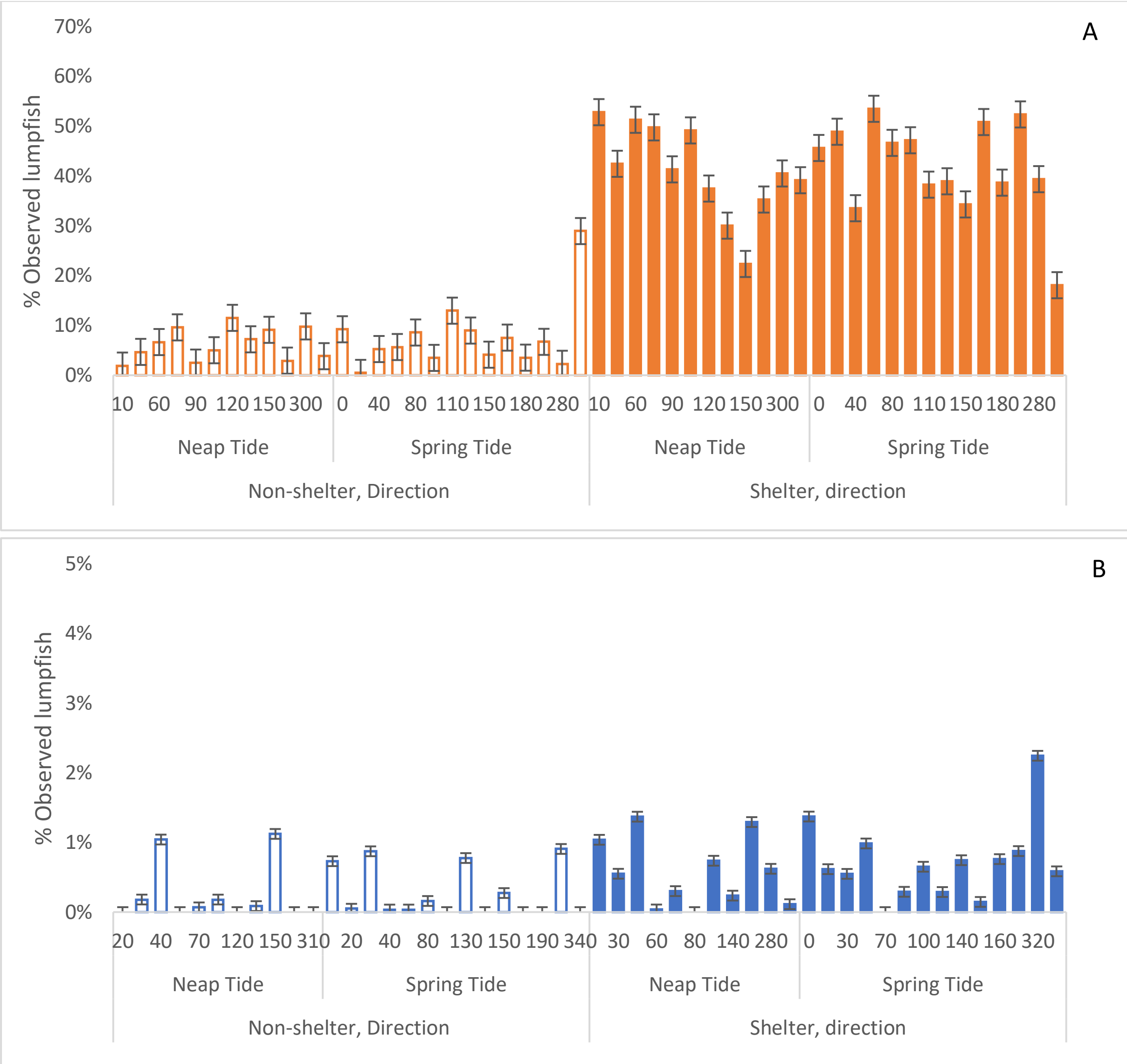

Figure 15: Mean observed lumpfish (± SE) at A) 3 m and B) 8 m depth, divided duration (min) the current entered the sector in Neap tide and Spring tide for group 1(NS=Non-shelter, S=Shelter). Note: different y axis range in A) and B).

3.3.1 Groupbased current affecting of lumpfish distribution

It was further interesting to test if correlation between measured maximum current (cm/s) and duration (min) the current entered the sector was different with shelter placement (Figure 7) and tidal changes (Figure 6). Therefore, it was set two different groups based on placement of the shelter. Cages 1 & 2 where group 1 and cages 10 & 11 were group 2.

3.3.1.1 Group 1

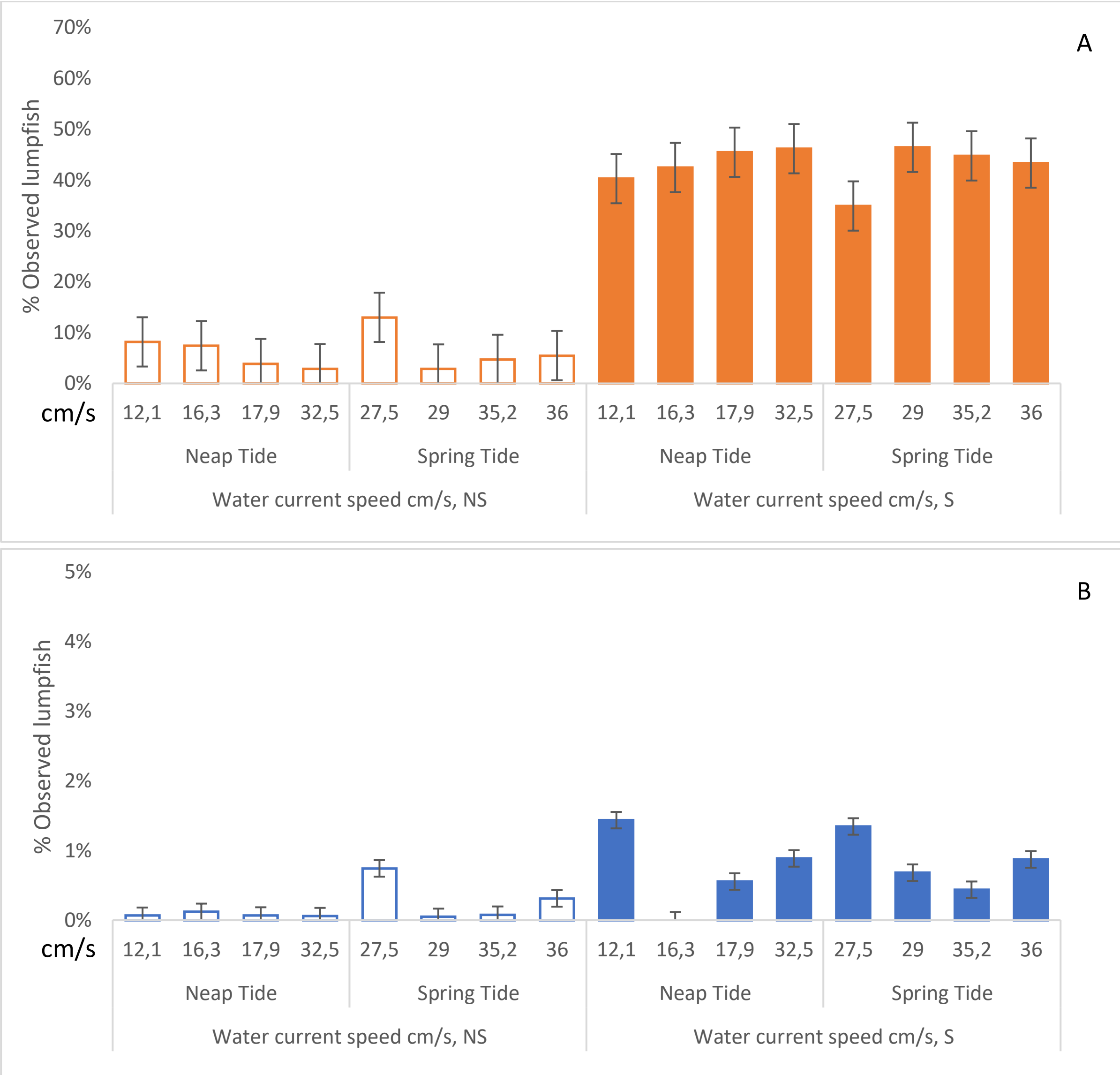

Figure 16: Mean % observed lumpfish (± SE) at A) 3 m and B) 8 m depth, at maximum measured current (indicated on X-axis in cm/s) during the observation period, during neap tide and spring tide and for different sectors (NS=Non-shelter, S=Shelter). Note: different y axis range in A) and B).

In total, during the entire period there is no correlation between water current speed and observed lumpfish at 3 and 8 m depth (-0.120, p=0.184; 0.128, p=0.200) (Figure 16). There is a non-correlation between duration (min) the current entered the sector and observed lumpfish at 3 and 8 m depth (0.09, p=0.306; -0.013, p=0.884) (Figure 17).

During spring tide, the correlation between observed lumpfish at 3 m depth and maximum measured current showed non correlation (-0.069, p=0.614) (Figure 16). Observed lumpfish and duration (min) the current entered the sector showed non correlation (-0.224, p=0.081) (Figure 17). At 8 m depth, observed lumpfish and measured current speed were negligible correlation (-0.184, p=0.203). duration (min) the current entered the sector also showed non correlation (0,002, p=0.986).

During neap tide, the correlation between observed lumpfish at 3 m depth and maximum measured current showed negligible correlation (0.69, p=0.614) (Figure 18). It was found negligible correlation with observed lumpfish and duration (min) the current entered the sector (-0.224, p=0.081) (Figure 19). At 8 m depth, observed lumpfish and measured current speed were non correlating (-0.184, p=0.203). Duration (min) the current entered the sector also showed non correlation (0.02, p=0.986).

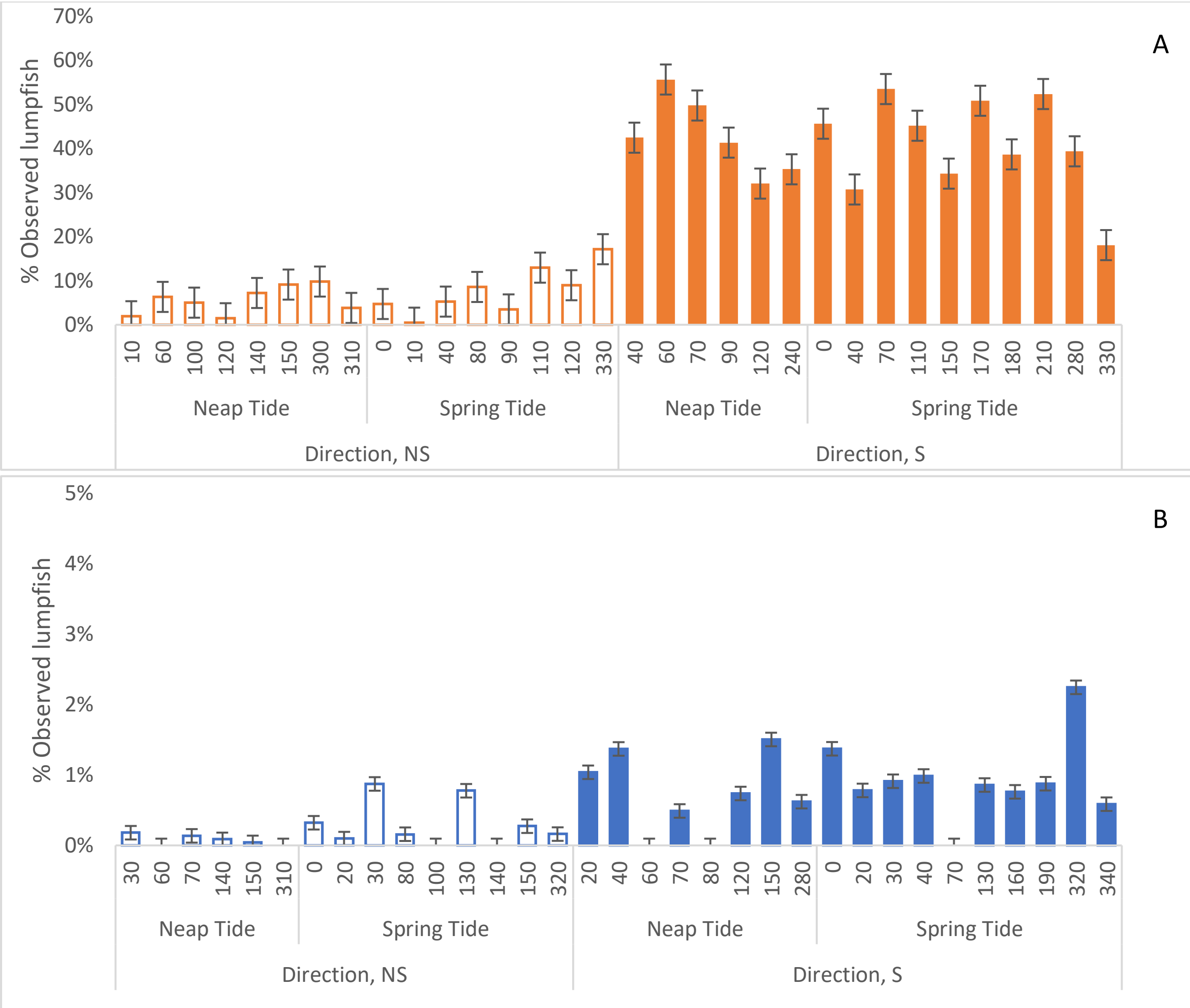

Figure 17: Mean observed lumpfish (± SE) at A) 3 m and B) 8 m depth, divided duration (min) the current entered the sector in Neap tide and Spring tide for group 1(NS=Non-shelter, S=Shelter). Note: different y axis range in A) and B).

3.3.1.2 Group 2

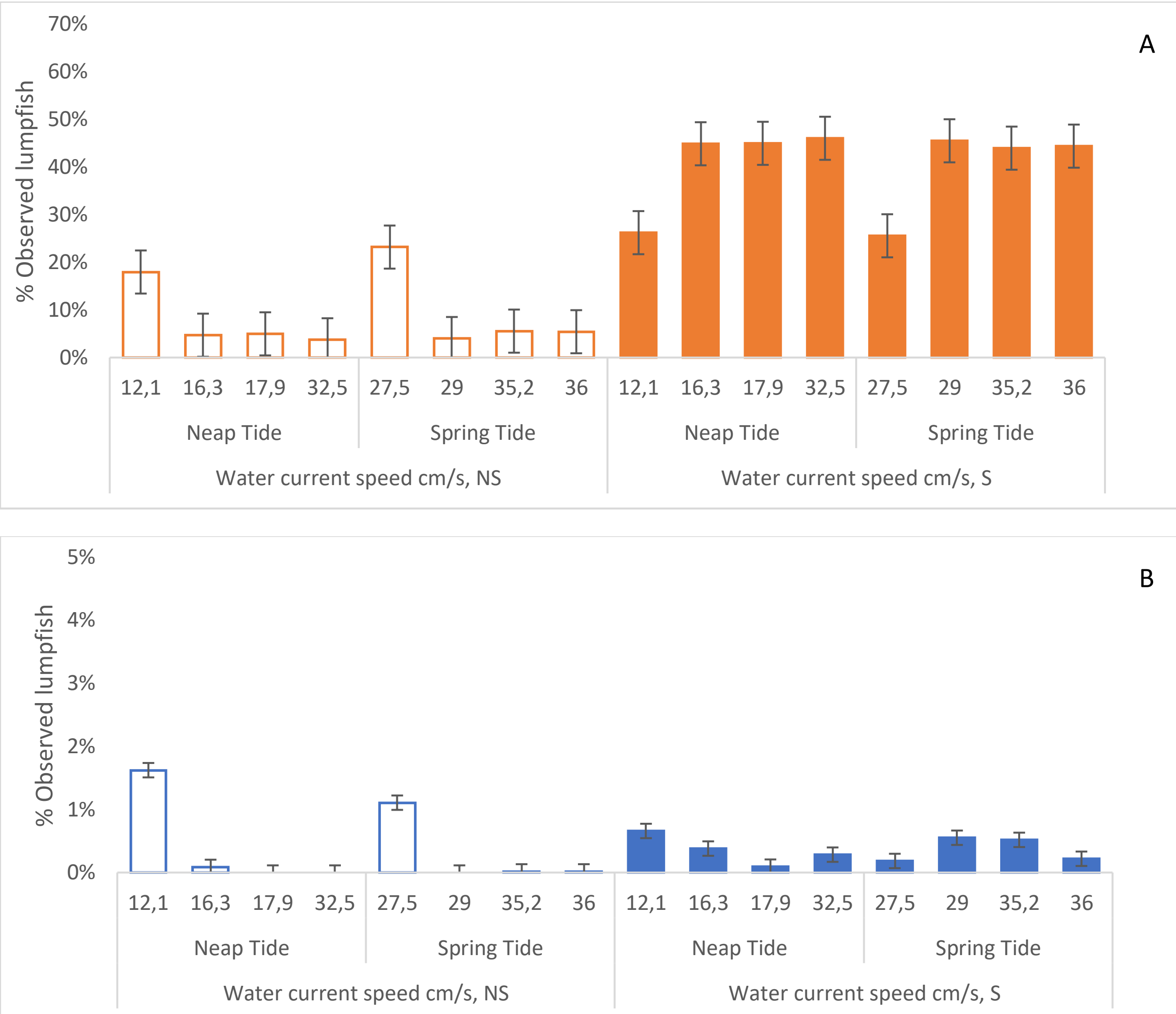

Figure 18: Mean % observed lumpfish (± SE) at A) 3 m and B) 8 m depth, at maximum measured current (indicated on X-axis in cm/s) during the observation period, during neap tide and spring tide and for different sectors (NS=Non-shelter, S=Shelter). Note: different y axis range in A) and B).

In total, during the entire period there is no correlation between water current speed and observed lumpfish at 3 and 8 m depth (-0.047, p=0.603; 0.187, p=0.052) (Figure 18). There is no correlation between duration (min) the current entered the sector and observed lumpfish at 3 and 8 m depth (0.082, p=0.349; -0.032, p=0.719) (Figure 19).

During spring tide, the correlation between observed lumpfish at 3 m depth and maximum measured current showed negligible correlation (0.088, p=0.523) (Figure 18). Low negative correlation with observed lumpfish and current in each minute for different sectors (0.323, p=0.011) (Figure 19). At 8 m depth, observed lumpfish and measured current speed were negligible correlation (0.348, p=0.015). Measured current in each minute for different sectors also showed negligible correlation (0.058, p=0.665).

During neap tide, the correlation between observed lumpfish at 3 m depth and maximum measured current showed no correlation (-0.193, p=0.158 (Figure 14). No correlation with observed lumpfish and duration (min) the current entered the sector (0.106, p=0.406) (Figure 15). At 8 m depth, observed lumpfish and measured current speed had negligible correlation (-0.025, p=0.869). Duration (min) the current entered the sector and observed lumpfish also showed non correlation (0,174, p=0.213).

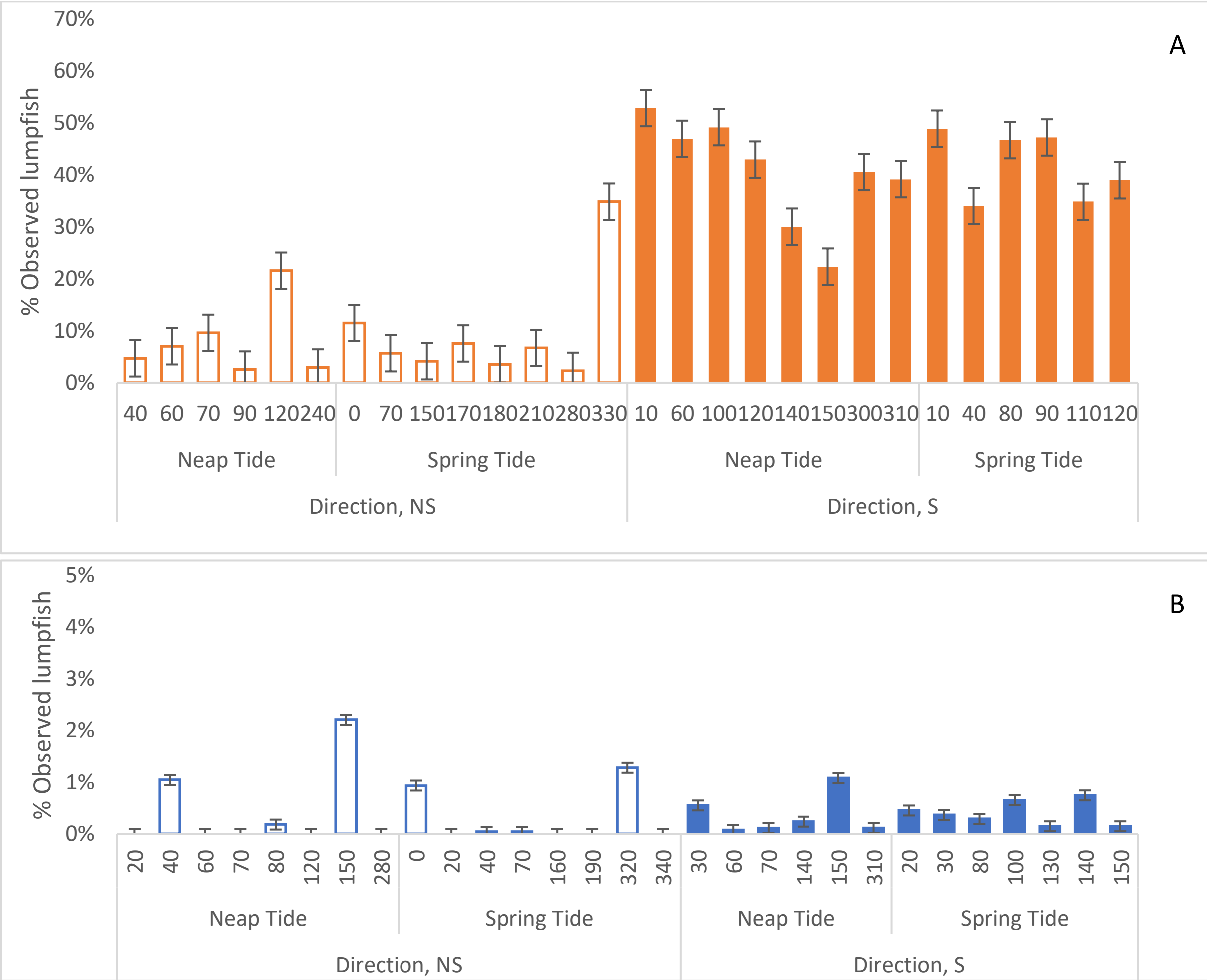

Figure 19: Mean observed lumpfish (± SE) at A) 3 m and B) 8 m depth, divided duration (min) the current entered the sector in Neap tide and Spring tide for group 1(NS=Non-shelter, S=Shelter). Note: different y axis range in A) and B).

4 Discussion

This thesis aimed to observe the lumpfish in a full-scale aquaculture farm and determine which factors are affecting lumpfish placement and distribution in shelter, at 3, and 8 m depth. The lumpfish was observed with an underwater drone during different tidal conditions. Different environmental sensors were used for data sampling.

4.1 Lumpfish distribution

Total observed lumpfish for the observation period showed significantly more lumpfish in 3m ($p=0.001$) compared to 8 m depth. This result is in line with other studies, where lumpfish activity in a cage has been observed. Lumpfish seem to be more distributed in the shallower parts of the cage Stenersen (2020), (Skiftesvik et al., 2018) also observed lumpfish in the upper part of the cage. There are not many studies of the vertical movement and depth distribution of the lumpfish under natural conditions. Kennedy et al. (2016) marked 41 lumpfish and the vertical movement was down to 418 m. Wild and farmed lumpfish cannot be compared, but we believe that if the cage had been deeper or the feeding had been adjusted to deeper parts of the cage, the lumpfish would have moved deeper as well (Skiftesvik et al., 2018).

There was significantly more lumpfish in the observation period in the shelters at 3 m depth, under all the different current conditions, than in the non-shelter sectors. At 8 m depth there was no significant difference between the sectors when it comes to number of lumpfish. Only the total of the 8 observations of the lumpfish was significant ($p=0.001$). These results are the same as Killen et al. (2007), who observed lumpfish in the shelter waiting for prey and not using the aerobic scope and their swimming capacity (Hvas et al., 2018). Skiftesvik et al. (2018) also observed more lumpfish in shelter than outside of the shelter. Skiftesvik et al. (2018) used PIT tags(*Passive Integrated Transponders*). In this study, an underwater drone was used, and the drone could potentially cause stress, increased swimming activity and hiding in shelters. If we have used PIT tag or a tracking system, our study would be more comparable to the other studies that have observed lumpfish in cages. With two sectors and the drone moving along the net, more lumpfishes could potentially be counted on the sides of the shelter compared to the center of the cage. An anecdote from the field is that the lumpfish was placed between shelter and the net pen. With more sector e.g., 60 sectors, the distribution of lumpfish in the cage would have been more detailed. Stenersen (2020) used different sectors and counted the number of lumpfish per minute. If the cage was divided in only 4 or 2 sectors, a more pronounced uncertainty would be introduced in the study.

A higher number of observed lumpfish at 3 m depth could also be a result of the water current and the reduction of water current using lice skirts (Frank et al., 2015). Since the lumpfish have thermal preference between 3-15 °C (Hvas et al., 2018) and the sea temperature did not go cross the preferred temperature, it would be interesting to observe the depth distribution at summer with higher temperatures. Feeding of the lumpfish was also done between the two shelters, and with a prey easily available, it could also affect the result with more lumpfish in shelter than outside. Skiftesvik et al. (2018) claimed that the lumpfish would preferer to be where it is easiest to get food. An anecdote from the observation: The lumpfish observed at 8 m depth seemed to be larger, more robust and seemed to tolerate a higher water current (Hvas et al., 2018). It was also this study observed larger lumpfish at 8m depth than in upper water layer. Since few lumpfish were observed at 8 m depth, it would be interesting to test whether they are in the center of the cage, where the Atlantic salmon is and more food from salmon pellets.

4.2 Tidal conditions affecting lumpfish distribution

All the observations summarized showed no correlation between observed lumpfish at 3m depth and horizontal water current speed and duration (min) the current entered the sector. Observed lumpfish only correlated under neap tide between measured current speed, with negligible correlation (0.252, <0.013) at 8 m depth.

When the cages were divided into 2 groups it was again observed low correlations. Under neap tide for group 2 it was negligible correlation at 3 m depth and duration (min) the current entered the sector (0.323, $p<0.011$). Also, at 8 m depth between observed lumpfish and current speed a negligible correlation was found (0.348 <0.015).

There are not many studies that have been testing different current conditions and lumpfish settlement. Jónsdóttir et al. (2019) classified aquaculture locations which exceeded the critical swimming speed for lumpfish, but did not observe lumpfish at the site. We have also measured current higher than the critical swimming speed (Hvas et al., 2018) for lumpfish. One reason why the lumpfish is not affected by the water current could be the use of lice skirt and the reduction *of current* in both net and lice skirt combined (Frank et al., 2015). It would be an improvement of this study to have cages both with and without lice skirt on the same aquaculture site. It would also be interesting to test out how much the bathymetry and site facilities reduce or increase the water current Jónsdóttir et al. (2021) have found a reduction of water current in the lice skirts of 20- 70 %, compared to outside the skirts. The current inside of the cage could also be reduced by fish movement and other factors. In this thesis we could not measure at two places at the same time due to large costs for two current sensors. We don’t know if the fish in the cage interrupt the measurement (Klebert and Su, 2020).

4.3 Evaluation and challenges with the methods

The observation of lumpfish was done at one fish farm, four cages on eight different days and video of 32 observations of lumpfish done with an underwater drone. The eight days were chosen based on different current conditions as neap and spring tide (Figure 6). It was not interrupted by any operations at the fish farm and the observation dates gave a good standardization and a large data set. There were no technological problems during the observations. The drone got stuck in the shelter under observation only a few times. Data analysing after each observation was time consuming due to the manual counting of lumpfish observed in the videos. If PIT tag or other tracking equipment had been used, time could have been saved. It was a strength in this thesis, from the beginning it was determined to standardize the routines for fieldwork and the analysis. We decided to use the total observed lumpfish rather than the estimate from the fish farm, due to uncertainties in numbers and mortality (Appendix, Table 1). This could have been done in a small-scale observation with several replicates, and the water current could be adjusted manually. Hvas et al. (2018) measured critical swimming speed in a swimming tunnel, and it would have been interesting to study the combination of a swimming tunnel with shelter, lice skirt and a net.

4.4 Further studies

This thesis has found and confirmed that lumpfish prefer to be at 3m depths compared to 8 m. Lice skirts improve the conditions for lumpfish and can be a tool at exposed sites with large water currents, where control method with cleaner fish is relevant. Further studies should test the distribution of lumpfish with and without lice skirts and how the current affect the placement of lumpfish. PIT tags or a tracking system would have allowed observations for longer time periods. In a future study, both salmon and lumpfish should have been observed simultaneously, to check for correlation in sectors and time, and to see if lice eating is based on interaction between the two species or if it just coincidence

In laboratory experiments, lumpfish distribution in a swimming tunnel with shelter and lice skirt, or shelter only, could have been studied. Further studies should also focus on depth distribution at different seasons to investigate if it is any seasonal changes. Our observations were done in late winter/early spring. We would also recommend looking for other seasonal changes. The result of this study can be used to optimize the conditions for the lumpfish and as a guide when removing lumpfish from the cage before delousing.

5 Conclusion

The results showed that lumpfish was significant more abundant at 3m depth than at 8 m depth ($p<0.05$). Lumpfish was also observed more in shelter sectors (28640 observed in shelter sector and 4529 observed in non-shelter sector). Lumpfish at 3 m depth was observed significantly more in the shelter than non-shelter sectors($p<0.05$). Lumpfish observed at 8 m depth was significantly more found in shelter than in non-shelter sectors ($p<0.05$). For spring and neap tide observations at 8 m depth, there were no differences in number of lumpfish in shelter and non-shelter. Lumpfish was not correlated with the different current conditions at neap tide and at spring tide for both depths. Use of lice skirts decreases the water current entering the cages, and this may have contributed to this result and seem to improve the environmental conditions for lumpfish in a commercial aquaculture cage. The result of the present study highlights the complexity of water currents and the distribution of lumpfish in a cage.

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Appendix

Appendix 1: Overview over observed lumpfish in the different cages for observation dates.

Table 1: Overview over observed lumpfish in the different cages for observation dates.

Cage	04.03	12.03	20.03	27.03	02.04	10.04	18.04	24.04
1	1886	1166	1039	934	1199	828	628	842
2	802	603	1864	1826	1553	1296	1283	1221
10	1090	1270	1374	1336	1527	1113	1374	889
11	379	48	612	843	673	714	751	878
Total	4137	3087	4889	4939	4952	3951	4036	3830

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