Caligus elongatus and *Lepeophtheirus salmonis* infestation on farmed and wild fish.

Master of Science in Marine Biology
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February 2022

Cover photo: Female *C. elongatus* with egg strings on Atlantic salmon, by Silje Marie Haugland Ryland.

Acknowledgments

This thesis was written in 2020-22 as a master's degree in marine biology at the Institute of bioscience at the University of Bergen. The scope of the task is 60 credits. Experiments and laboratory work have been carried out at the Institute of Marine Research (IMR) in Bergen, Matre, and Austevoll.

Firstly, I want to thank my brilliant principal supervisor, Sussie Dalvin, for allowing me to work on this project and guiding me throughout the process. I am very grateful for the close follow-up and helpful advice on my assignment. I appreciate all the knowledge shared and all the discussions we have had over the past year. Furthermore, I would like to thank my co-supervisor, Ørjan Karlsen, for helping me collect material in the field, and Egil Karlsbakk for good advice and feedback on the thesis and help with statistical analysis. Stig Mæhle has guided me with the molecular biology challenges I have had along the way and given me valuable laboratory training. Your door has always been open, and when you did not have time, you made some. I would also thank the staff at IMR for their assistance in the louse collection for my project.

The task would not have been possible without the Institute of Marine Research. An extra thank you to the research group of pathogens and disease transfer who financed the project, allowing me to participate in a course in species identification, stage determination, and counting lice on salmonids. I have gained a much broader lab experience simultaneously as I participated in the field.

It has been a fascinating period being part of, and included, in such a skilled and knowledgeable research group while working on my thesis. I feel very fortunate. All of you have been supportive, motivating, and understanding. I have learned many different biological methods which may be helpful in the future. I want to thank all of you for your patience during the work period.

Finally, I wish to thank my life partner, Jens, my sons Aksel and Elias, and last but not least, the rest of my family and friends for all the support, patience, and love during this period. Thank you.

Bergen, February 2022

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Abstract

The sea lice *Caligus elongatus* and *Lepeophtheirus salmonis* are important parasites infecting wild and cultured salmonids. Due to the extensive problems and costs the sea lice cause, it is important to understand their biological processes and pathology to develop better methods for controlling the parasites. Previous research has mainly focused on *L. salmonis* (the salmon louse) and all legislation around sea lice at fish farms in Norway is aimed at it. The abundance of *C. elongatus* has increased in the last decades and gaining knowledge about *C. elongatus* fecundity is important in order to understand the dynamics of the sea louse infestations. The sea lice species, *C. elongatus,* and *L. salmonis*, are challenging to distinguish from another in the early life stages. A method to separate them macroscopically would make it easier for fish farmers to report the correct abundance of *L. salmonis*. Furthermore, identifying previous hosts of *C. elongatus* would make us able to understand the movements of the lice.

Lice were collected from different locations along the Norwegian coast and were used to study the fecundity of *C. elongatus*. An infection experiment of *C. elongatus* and *L. salmonis* on Atlantic salmon (*Salmo salar*) was performed to examine if there are preferred attachment sites on the fish in the chalimi stages. Secondly, the experiment examined to what extent chalimi *C. elongatus* and *L. salmonis* can be distinguished by macroscopical examination based on the characteristics made from previous research. The sea lice species was assumed, and a PCR and agarose gel electrophoresis confirmed the correct species. Finally, an experiment was performed to investigate if the previous host of *C. elongatus* could be identified. The sea lice feed on the host's skin, and molecular analysis of the sea lice's DNA might detect the fish's DNA in the gut content of the louse. Knowledge of the previous host of *C. elongatus* can contribute to identifying the source of *C. elongatus* infections at fish farms.

The results of *C. elongatus* fecundity showed that the origin of the host had a significant impact on the lice´s size and the number of eggs, whereas lice from wild fish were larger and had more eggs. There was a positive correlation between egg string length and the number of eggs. There was no correlation between the lice length and the number of eggs. *Caligus elongatus* showed a significant difference in the number of eggs from the different regions in Norway. The host specie affects the number of eggs, but not the length of the lice. The attachment sites of the sea lice were similar for both sampling groups, where the dorsal fin was the predominant location, followed by the posterior back. Macroscopical identification of the chalimi stages of the sea lice species was mainly based on the body- and eye pigmentation. The characteristics used were challenging to observe macroscopically, and therefore, it is not possible to successfully identify

previous hosts using the characteristics as described in this thesis. Investigation of the attachment sites and macroscopical identification was performed simultaneously with the same material. The infection rate of *C. elongatus* was very low and it was not possible to investigate a preferred attachment pattern of *C. elongatus*, which also affected the macroscopical identification experiment. Identification of the previous host of *C. elongatus*, showed that 2 out of 10 samples were correctly identified by the method. The study identified DNA from a previous host in the gut content of *C. elongatus*. In addition, the saithe experiment found DNA from the fish host in the gut content of the lice after 22 hours.

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I. Introduction

1.1 Sea lice

Sea lice are parasites living on (ectoparasite) and obtaining nutrients from their host. They have a direct lifecycle, meaning they only need one host to complete their life cycle (Kabata, 1979; Wootten, Smith, & Needham, 1982). The symbiotic relationship between a parasite and its host is often described as a coevolutionary arms race where they have coexisted for a long period, and the only way for the host to defend itself is to avoid the parasite (Bui, Oppedal, Stien, & Dempster, 2016; Sukhdeo & Moore, 2002). The sea lice species *Caligus elongatus* (Nordmann, 1832) and *Lepeophtheirus salmonis* (Krøyer, 1837) are common, marine parasites that represent a threat to wild, and farmed fish in the northern hemisphere, whereas *C. elongatus* can be found in both hemispheres. Both species belong to the subphylum Crustacea, subclass Copepoda, order Siphonostomatoida, and the family Caligidae where all species are parasitic with a flattened body and appendages to attach to their host (Wootten et al., 1982).

Nordmann (1832) was the first to make a description of *C. elongatus*, Parker (1969) made an improved description. Later, descriptions and illustrations of *C. elongatus* developmental stages were made by Piasecki (1996). From here, several researchers worked on a description of the life cycle and the morphology of *C. elongatus* (Hogans & Trudeau, 1989a, 1989b; Piasecki, 1996; Piasecki & MacKinnon, 1993, 1995; Pike, Mordue, & Ritchie, 1993). Although *C. elongatus* is considered one species, there are two genotypes, genotypes 1 and 2 (Øines & Heuch, 2005). A study of the distribution of the genotypes of *C. elongatus* found that lice from northern Norway were genotype 1, while genotype 2 predominantly was found in southern Norway. No genotype 2 lice were found from the northernmost areas (Altafjorden and Sørøya) and genotype 2 lice were found from southern Norway (Karmøy, Hidra, and Frøya) which were collected at the same time of year as the lice from the current study (Øines & Heuch, 2007). This might indicate that there could be a north-south gradient of *C. elongatus* different genotypes.

Bishop Erik L. Pontoppidan (1698-1764) was the first to describe *L. salmonis*, followed by Krøyer (1837), who made a scientific description of the louse. Johannessen (1978) studied the early life stages of *L. salmonis,* but a complete description of copepodids to adults was not made until 13 years later by Johnson and Albright (1991b) and Scram (1993). Subsequently, Hamre et al. (2013) corrected the number of life stages. Pacific and Atlantic *L. salmonis* have co-evolved with different salmonids, and isolation has led to the development of two separate subspecies in the two oceans (Skern-Mauritzen, Torrissen, & Glover, 2014).

1.2 Aquaculture and control of sea lice

The growth of the aquaculture industry in Norway started in the 1960s and led to an increase of sea lice due to the high fish biomasses, i.e., host in open net-pen farms (Pike, 1989; Wootten et al., 1982). The Scottish farms were first affected by *C. elongatus*, but *L. salmonis* subsequently became the main problem (Pike, 1989); *C. elongatus* has therefore been referred to as 'the Scottish lice' in Norway. The sea louse *L. salmonis* is commonly called 'the salmon lice' due to its preference for various salmonids (Pike & Wadsworth, 1999). In Norwegian waters, *L. salmonis* has more impact on farmed than wild fish. In recent years, *C. elongatus* has started to cause problems at fish farms in parts of northern Norway (Hemmingsen et al., 2020). When production levels began to rise in the middle of the 1970s, there was an increase in epizootic outbreaks. This led to extensive and costly delousing with high mortality rates, which the aquaculture farmers have struggled with ever since (Heuch & Mo, 2001; Pike & Wadsworth, 1999; Torrissen et al., 2013). The costs to control the sea lice infestations in Norway were estimated to be 1 billion NOK in 2006 (Costello, 2009), and it was estimated that the parasitic lice caused Norway a total of 3.8 billion NOK in 2011 (Abolofia, Asche, & Wilen, 2017). A high abundance of sea lice is harmful to the fish welfare of farmed- and wild fish stocks. The parasites graze mucus, tissue, and blood from the host and thereby inflict wounds on the fish, which can cause secondary bacterial infections (Costello, 1993). Host responses to an infestation of sea lice are oedematous and hemorrhaged skin with abrasions where the lice have grazed (Wootten et al., 1982). In addition, osmoregulation problems by leakages, elevated stress levels, and weakened immune systems are common (Nolan, Reilly, & Wendelaar Bonga, 1999). *Lepeophtheirus salmonis* cause more extensive damage than *C. elongatus* (Pike & Wadsworth, 1999).

It is crucial to prevent and control the sea lice to protect wild and farmed salmon stocks and decrease economic costs in the aquaculture. The high abundance of *L. salmonis*, and the issues associated with the lice led to requirements to control the infestations of the lice at fish farms. There are strict obligations to report the abundance of *L. salmonis* to the Norwegian Food Safety Authority during weekly sea lice counts at Norwegian fish farms. Treatments are required if the number of lice exceeds specific criteria (Heuch & Schram, 1999; Ministry of Trade Industry and Fisheries, 2012). All legislation of sea lice in the farming industry in Norway is directed at *L. salmonis*, and there are no requirements to report the abundance of *C. elongatus* in the Norwegian sea lice regulations. There are no public registers of the actual number of *C. elongatus,* and it is difficult to estimate how severe the problem is. However, *C.*

elongatus has become so inconvenient that treatments are carried out against the lice in northern Norway with Emamectin benzoate and Slice (Imsland et al., 2019).

As *C. elongatus* and *L. salmonis* often occur simultaneously (Wootten et al., 1982), they need to be correctly distinguished from another to report the correct abundance of *L. salmonis*. Adult *C. elongatus* and *L. salmonis* can easily be distinguished morphologically but separating the chalimi larvae is challenging as the differences are less obvious. The similarities between the sea lice species in the chalimi stages might lead to incorrect identification of the species based on macroscopical investigations. A method to distinguish the chalimi stages of *C. elongatus* and *L. salmonis* might help separate the species morphologically.

More efficient and accurate methods to control the sea lice depend on detailed information about the lice distribution, abundance, and behavior (e.g., automated sea lice counting) (Bui, Oppedal, Nola, & Barrett, 2020). Current routines for monitoring *L. salmonis* at fish farms are time-consuming and consist of physically capturing and counting the number of lice on the fish. Salmonid hosts are equally susceptible to infestation by both sea lice species, which frequently are found on the same host, but it is common to find a greater number of *L. salmonis* than *C. elongatus* (Berland, 1993; Pike & Wadsworth, 1999; Todd, Whyte, MacLean, & Walker, 2006; Wootten et al., 1982). Preferred attachment sites of *C. elongatus* and *L. salmonis* on Atlantic salmon may improve the accuracy of sea lice monitoring on the fish. Previous studies have examined the attachment preference sites of *L. salmonis* and *C. elongatus*, but there are few reports of the attachment sites of chalimus *C. elongatus*.

1.3 Lifecycle and developmental stages

Caligus elongatus:

The lifecycle of *C. elongatus* is divided into eight developmental stages: the nauplii (I-II), copepodid, chalimi (I-IV), and the adult stage, where each stage is separated by a molt (ecdysis) (Figure 1). This is a process where the lice produce a new and larger exoskeleton underneath their old one and release their old exoskeleton in order to expand and grow (Eichner, Hamre, & Nilsen, 2014). The lifecycle starts with the adult female's two uniseriate egg strings extruded from the genital segment. Nauplius I larvae hatch directly from the egg strings into the water column, free-swimming with their three pairs of appendages. However, they can be dispersed over greater distances by coastal currents (Piasecki & MacKinnon, 1995). The nauplii larvae are oval, almost translucent, with a few dark brown pigments (Piasecki, 1996). They depend on the yolk sac's energy reserves for nutrients in the nauplii and copepodid stages until they attach

a host (Pike & Wadsworth, 1999). The larvae are positively phototactic and position themselves in the upper water layers to increase their possibilities of meeting a host swimming past (Hogans & Trudeau, 1989b)

The copepodid has a more streamlined shape than the nauplii larvae (Hogans & Trudeau, 1989b). The sea lice actively seek an appropriate host to attach in order to obtain nutrients and proceed the development. The copepodid uses its maxillipeds to grip the fish initially. If the host is the correct species, the lice anchor themselves to the scales or fin rays of the fish with a frontal filament that ensures permanent attachment to the host, restricting movement on the surface of the fish, hence the feeding area (Piasecki & MacKinnon, 1995). The frontal filament is formed in the cephalothorax as late copepodites. The slender frontal filament helps the lice to remain attached to the host during ecdysis and is merely extended at each ecdysis from chalimi I-IV (Piasecki & MacKinnon, 1993).

The shape of the chalimi larvae is more extended and broadened as they develop to chalimus IV (Pike, Rowand, & Mackenzie, 1993). At the same time, the body segmentation becomes more prominent. The chalimus larvae develop a shaper tip on the anterior part of the cephalothorax and the abdomen is half the length or as long as the cephalothorax in these stages (Piasecki, 1996). It is possible to distinguish the sexes from each other when the sea lice molt into the chalimus III stage, where the male´s abdomen is separated into two segments while the females consist of only one (Piasecki, 1996; Piasecki & MacKinnon, 1993, 1995). Copepodites and the chalimi stages are called the sessile stages due to the lice´s immobility.

The parasite detaches from the temporary frontal filament and can move around the fish's surface to graze in the mobile phase as adults (Wootten et al., 1982). Some adults remain in the same position where the frontal filament has been their whole life (Piasecki & MacKinnon, 1995). Adult *C. elongatus* develops characteristic lunules in the front of the cephalothorax that acts as a suction cup against the host. Species in the genus *Caligus* develop such lunules (Hogans & Trudeau, 1989b; Kaji et al., 2012), and this characteristic can be used to separate the *Caligus* species from *L. salmonis*. Adults develop a typical yellow-brown body-color (Hogans & Trudeau, 1989b).

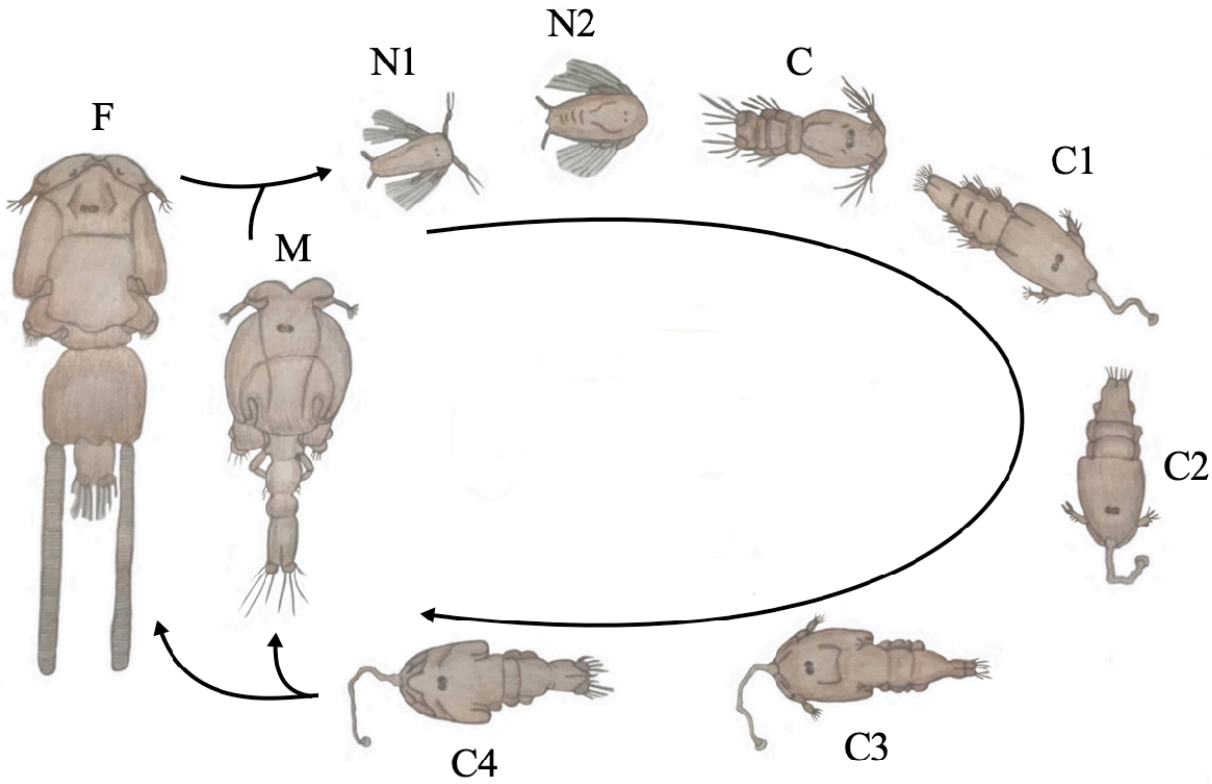

Figure 1. *Drawing of C. elongatus lifecycle. Nauplius I (N1), nauplius II (N2), copepodid (C), chalimus I (C1), chalimus II (C2), chalimus III (C3), chalimus IV (C4), adult male (M) and an adult female with egg strings (F). Figure by author inspired by Piasecki (1996).*

Lepeophtheirus salmonis:

The life cycle of *L. salmonis* consists of eight stages, they have two pre-adult stages (I-II) which substitutes the chalimi stages (III-IV) of *C. elongatus*; the nauplii (I-II), copepodid, chalimi (I-II), pre-adult (I-II), and the adult stage (Figure 2) (Hamre et al., 2013). The body shape from nauplius to chalimi larvae is similar to that of *C. elongatus*, with an elongated oval shape, but *L. salmonis* larvae are larger. The copepodid has a light-brown body color and develops scattered brown spots in the chalimius stages (Schram, 1993). Chalimus II larvae develops an unpigmented area around the eyespots. It is possible to distinguish the sexes morphologically in the late chalimus II stage, where the females have a longer cephalothorax than males (Eichner et al., 2014). However, it is more prominent when the females get a triangular-shaped genital segment while the male segment is barrel-shaped as pre-adult I (Schram, 1993). As opposed to *C. elongatus,* the frontal filament of *L. salmonis* are short, thick, and are not extended at each ecdysis but renewed at each molt in the sessile phase until they become mobile as adults (Gonzalez-Alanis, Wright, Johnson, & Burka, 2001; Pike, Rowand, et

al., 1993; Pike & Wadsworth, 1999). *Lepeophtheirus salmonis* lack the characteristic lunules the *Caligus* species develop as adults (Hogans & Trudeau, 1989b; Kaji et al., 2012).

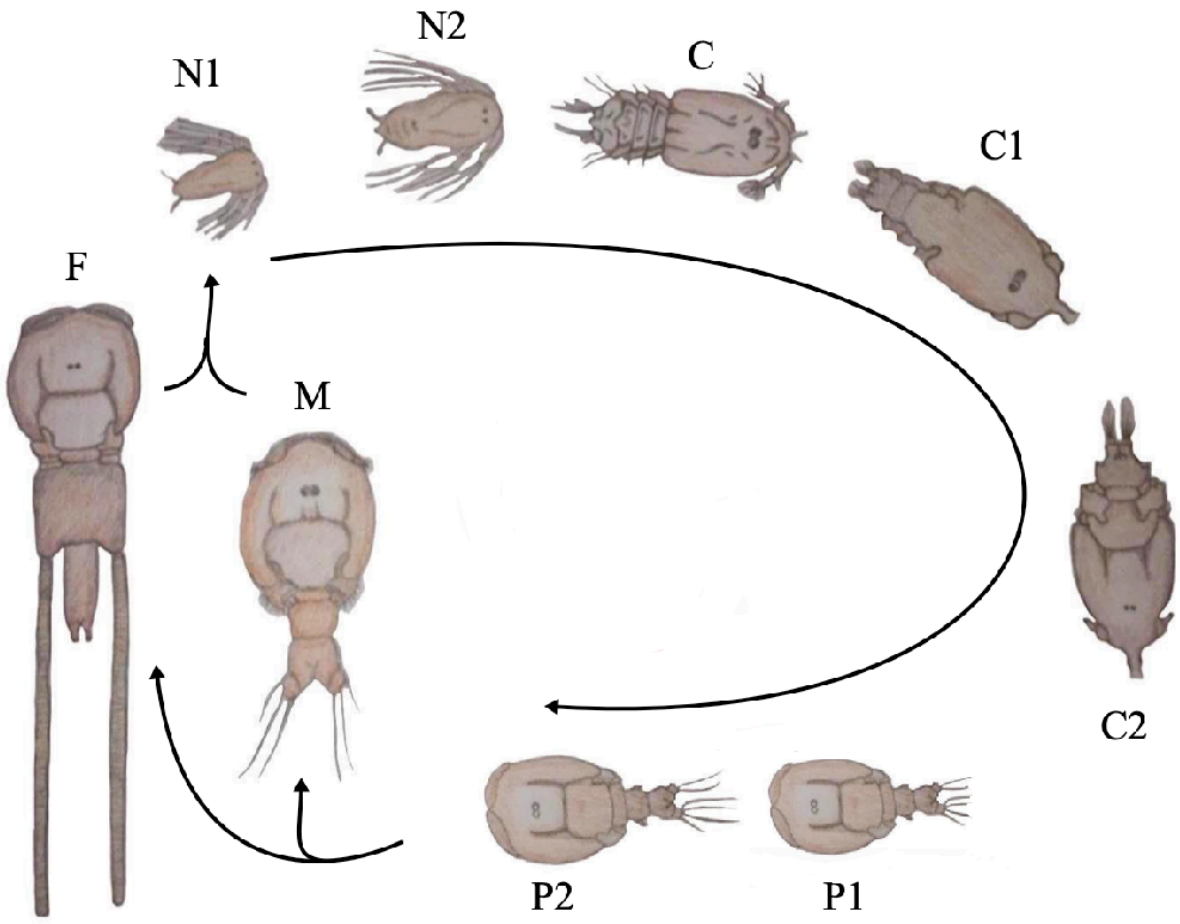

Figure 2. *Drawing of L. salmonis life cycle. Nauplius I (N1), nauplius II (N2), copepodid (C), chalimus I (C1), chalimus II (C2), pre-adult I (P1), pre-adult II (P2), adult male (M) and an adult female with egg strings (F). Figure by author inspired by Schram (1993).*

1.4 Reproduction

Both *C. elongatus* and *L. salmonis* have internal fertilization and an oviparous reproductive strategy (Crawford, Dill, Finstad, Todd, & Fraser, 2009). They are poikilotherms, and temperature is therefore of great importance for reproductive output and developmental rate (Nordhagen, Heuch, & Schram, 2000). Both sea lice species are present all year, and gravid females occur at all times (Wootten et al., 1982). Mating starts with a male searching for a female on the fish, as the males become mobile before the females. Adult males of *C. elongatus* (Piasecki & MacKinnon, 1995) and *L. salmonis* (Ritchie, Luntz, Pike, & Rae, 1996) may grab a pre-adult female still attached with their frontal filament, and wait for her to molt and become sexually mature. This pair is called “precopula”, and the male’s behavior is called mate

guarding (Boxshall, 1990). Female's store sperm received from the males in a sperm receptacle called spermatophore and releases sperm to fertilize the eggs as they are pushed into the egg strings. The eggs mature in the genital segments before being packed into the egg sac (Dalvin et al., 2011). The eggs are disc-shaped and are carried in two cylindrical strings attached to the female genital segment. The eggs are uncolored at first and darken with maturation due to the development of embryo pigments. The eggs hatch in sequence from the end of the egg string towards the female genital segment (Hogans & Trudeau, 1989b; Wootten et al., 1982). A few hours after the eggs have hatched, the female lice produce new egg strings with eggs that lie ready in the genital segment (Piasecki & MacKinnon, 1993). The females continuously produce new eggs even if they are not fertilized (Eichner et al., 2008; Nordhagen et al., 2000).

The generation time (newly hatched nauplius larvae to mature adults) for *C. elongatus* is approximately 6.2 weeks at a temperature of 10 °C. Nauplius I lasts 24 hours before ecdysis to nauplius II, which lasts for 67 hours at 10 °C (Piasecki & MacKinnon, 1995; Pike, Mordue, et al., 1993). *Caligus elongatus* produces at least two sets of egg strings (Piasecki & MacKinnon, 1995), but there are few studies on the number of eggs produced in the egg strings of *C. elongatus*. However, Hogans and Trudeau (1989a) found 89 eggs in each egg string, Pike et al., (1993) found 80 eggs in each egg string, and Jackson and Minchin (1992) observed 54 and 89 eggs per egg string, respectively.

According to Albright and Johnson (1991), the generation time for *L. salmonis* is 7-8 weeks at a temperature of 10 °C, while Hamre et al. (2019) found a generation time of 5.7 weeks at 9°C. There have been reported substantial differences in the size of *L. salmonis* depending on their host origin. Factors that affect the body size of the sea lice are temperature (S. Dalvin personal communication), origin, and the year's season (Pike & Wadsworth, 1999). The first set of egg strings is shorter than all subsequent egg strings. Adult *L. salmonis* females produce at least 11 pairs of egg strings with thousands of eggs during their lifecycle (Nordhagen et al., 2000). Hamre, Glover & Nilsen (2009) observed 15.5 months old *L. salmonis* females still reproducing under laboratory conditions. The number of eggs varies from 100 to 1000 eggs pr. egg string (Costello, 1993), but lice from farmed Atlantic salmon (*Salmo salar*) produce, on average, about 200 eggs per egg string (Brooker, Skern-Mauritzen, & Bron, 2018). The egg sacs may be more than twice their body length, up to 20 mm, but this varies considerably (Pike & Wadsworth, 1999; Wootten et al., 1982).

Detailed descriptions of the fecundity and the developmental rate of *C. elongatus* might facilitate strategies to prevent the increased abundances of *C. elongatus* at Norwegian fish farms.

1.5 Host specificity

Caligus elongatus is a common parasite in the North Atlantic Ocean with a low host specificity, the lice has been collected from more than 80 different marine fish species (Costello, 2006; Kabata, 1979). Different studies have shown that *C. elongatus* in Norwegian waters are particularly associated with lumpfish (*Cyclopterus lumpus*), pollock (*Pollachius pollachius*), sea trout *(Salmo trutta)*, herring (*Clupea harengus*), saithe (*Pollachius virens*), and cod (*Gadus morhua*) as hosts (Boxshall, 1974; Heuch, Øines, Knutsen, & Schram, 2007; Øines, Simonsen, Knutsen, & Heuch, 2006). *Caligus elongatus* is considered a better swimmer than *L. salmonis,* and it can transfer among hosts as a natural part of its life strategy leading to sudden large populations of sea lice on fish not previously infected (Hogans & Trudeau, 1989a; Pike & Wadsworth, 1999). It is speculated where the sudden infection of the sea lice at fish farms comes from, but it is assumed that infestations of *C. elongatus* at fish farms have been connected to passing schools of pollock, saithe, and herring (á Nordi et al., 2015; Hemmingsen et al., 2020). Adult *C. elongatus* unattached from the original host can re-infect other fish species, which may explain the rapid increase of *C. elongatus* in fish farms (Heuch et al., 2007). Previous hosts of *C. elongatus* found on fish farms might indicate the source of infestations at fish farms and help us one step closer to controlling and monitoring the sea lice.

Lepoptherius salmonis is a host-specific parasite, specialized and restricted to salmonid fishes of the genera *Salmo, Salvelinus, Oncorhynchus,* and *Coregonus* in the northern hemisphere (Kabata, 1979). In Norway, this includes the native Atlantic salmon*,* Arctic char *(Salvelinus alpinus),* and sea trout (Hamre, Bui, Oppedal, Skern-Mauritzen, & Dalvin, 2019; Pike & Wadsworth, 1999). *Lepeophtheirus salmonis* often spend the entire life on the same host it first attaches to (Wootten et al., 1982).

1.6 Aims of study

More knowledge of sea lice's biology and reproductive potential is of great interest to contribute to developing methods to prevent and control the lice as it is an important challenge for the aquaculture industry. Specific morphological characteristics to separate the sea lice species and preferred attachment sites would make it easier and more efficient at sea lice monitoring for fish farmers. Knowledge of previous hosts of *C. elongatus* might contribute to identifying the source of sea lice infestations at fish farms, which might be important to fight the parasite and handle it. A core element to resolve the issue is to gather more information about the epizootiology and perform more research on the given area. The present study investigated the occurrence of sea lice on wild and farmed Atlantic salmon and quantified differences between the lice species. Consequently, the aims of this study were to:

(1) Study the fecundity of *C. elongatus* as measured by egg number and size of egg strings.
(2) Examine if chalimi *C. elongatus* and *L. salmonis* have preferred attachment sites on Atlantic salmon.
(3) Examine to what extent chalimi *C. elongatus* and *L. salmonis* can be distinguished by macroscopical examination.
(4) Test if it is possible to identify previous hosts of *C. elongatus* by molecular analyses on gut content.

2 Materials and method

2.1 Collection of C. *elongatus*

Caligus elongatus from wild fish were collected by the Institute of Marine Research during the annual salmon lice surveillance program along the Norwegian coast from May to September 2019 (Nilsen et al., 2019). The lice were put in 1.5 mL microtubes filled with 98% ethanol. The date, geographic location (Figure 3), and host species were recorded on the sample. *Caligus elongatus* from farmed fish was collected at Låva in Boknafjord, Rogaland, at an Atlantic salmon fish farm, and the lice were stored the same way as lice from wild fish. The sea lice collected are divided into different regions of northern Norway, central Norway, and southern Norway due to the discovery of genotype 1 in northern and genotype 2 in southern Norway (Øines & Heuch, 2007). Southern Norway consisted of 260 lice, of which 81 were from farmed salmon. Central Norway consisted of 6, and northern Norway had 22 lice (Table 1). A complete list of chemicals, primers, kits, instruments, and software used during this thesis are listed in Appendix A – Table 1-5A.

Table 1. *The site, location code, region, and decimal coordinates (latitude, longitude) of the collected lice along the Norwegian coast. Site A-K represents locations where lice from wild fish were collected, and the site marked with a star (*) represents the only location where lice from farmed fish were collected.*

Sea lice collection from wild and farmed fish			
Site	**Location**	**Region**	**Coordinates**
Bugøyfjorden, Troms and Finnmark	A	North	69.8670, 29.3900
Porsangerfjorden, Troms and Finnmark	B	North	70.5687, 25.4755
Altafjorden, Troms and Finnmark	C	North	70.1332, 23.0853
Reisafjorden, Troms and Finnmark	D	North	69.9404, 21.1562
Ullsfjorden, Troms and Finnmark	E	North	69.6572, 19.7677
Øksfjorden, Nordland	F	North	68.3692, 15.2988
Blindalsfjorden, Nordland	G	Central	65.1933, 12.2912
Namsenfjorden, Trøndelag	H	Central	64.4619, 11.9217
Nordfjorden, Vestland	I	South	61.8617, 6.0159
Sognefjorden, Vestland	J	South	61.1545, 6.5806
Boknafjorden, Rogaland	K*	South	59.3011, 6.3254

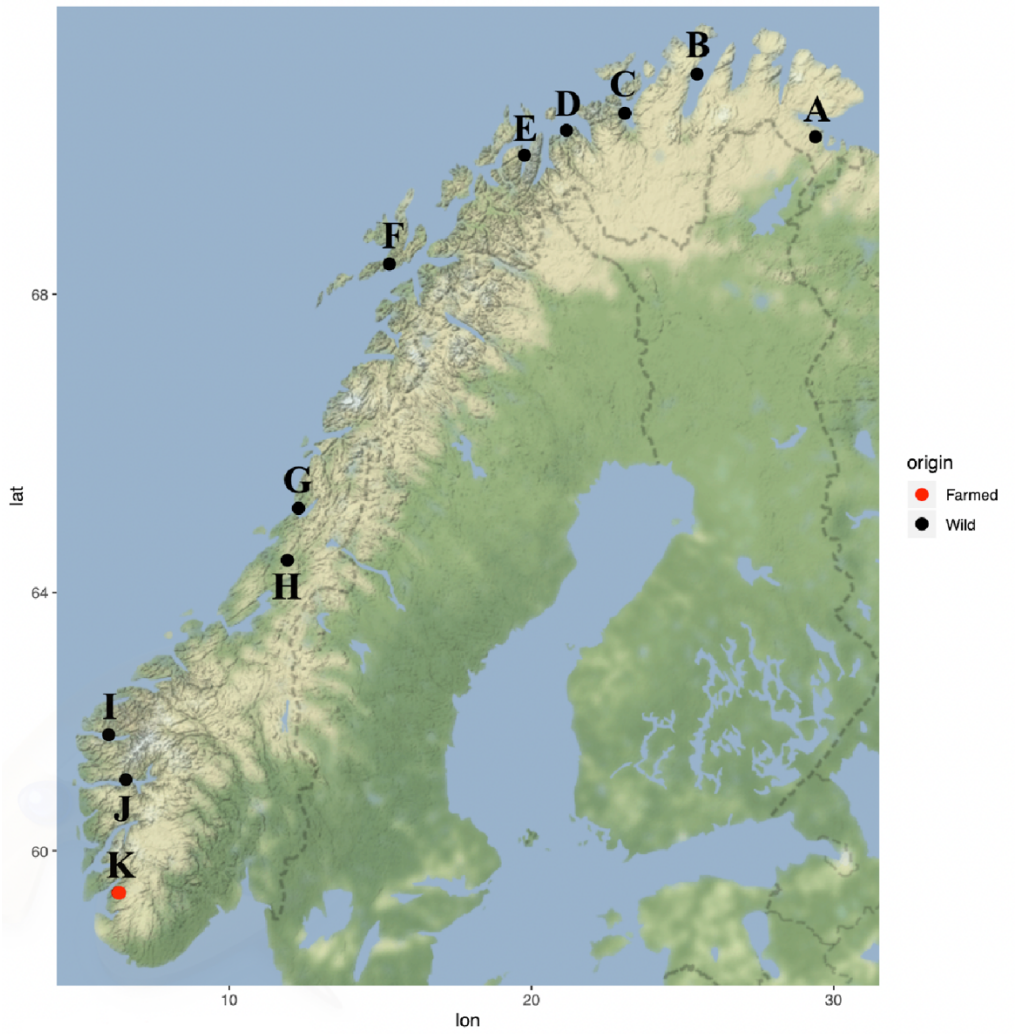

Figure 3. *A map of the Norwegian coast with associated longitude (lon) and latitude (lat) and dots that shows the locations from which lice were collected (Table 1). Bugøyfjorden (A), Porsangerfjorden (B), Altafjorden (C), Reisafjorden (D), Ullsfjorden (E), Øksfjorden (F), Blindalsfjorden (G), Namsenfjorden (H), Nordfjorden (I), Sognefjorden (J), Boknafjorden and Låva (K). The dots represent the origin of the host of which lice from wild fish are black, and lice from farmed fish are blue.*

2.2 Investigation of *C. elongatus* fecundity

Each female louse was individually examined in a petri dish filled with 96% ethanol. The lice were photographed using a stereomicroscope (Olympus SZ1). Morphometrics of the lice was measured by using the software ImageJ version 1.8.0 (https://imagej.nih.gov/ij/), which measures the exact length of the body and the egg strings of the sea lice on the pictures by calibrating ImageJ (Schneider, Rasband, & Eliceiri, 2012) (Figure 4). The images were also used to count the number of eggs inside each egg strand of the females. Only complete egg strings were included, meaning lice with egg strings that had started to hatch or were damaged were excluded. A total of 576 single egg strings were examined.

***Figure 4**. An illustration of the morphometric measurement lines of the body length (red) and the right egg string (blue) of a female C. elongatus photographed by a stereomicroscope.*

2.2.1 Duration of *C. elongatus* nauplius stages

The duration of *C. elongatus* two nauplius stages was examined for ten days in a study conducted in December 2019. The sea lice were provided by the Sea Lice Research Center (SLRC) at the University of Bergen (UoB) (genotype 1). *Caligus elongatus* leaves behind an empty exoskeleton at each molt. Therefore, the exoskeleton was used to indicate when the sea lice had molted into the next developmental stage. The days post hatching (DPH) until the first molt was used to estimate the duration of the nauplius I stage (M1). The days post nauplius I until copepodites were used to estimate the duration of the nauplius II (M2) stage.

The setup of the incubation system consisted of two boxes with 16 cylindrical wells each (Sea Lice Research Centre, 2020). The bottom of the wells was made of a thin sieve, and when a well was lifted, the water passed through the filter, and only the content was left in the well. Each of the 32 wells inside the boxes contained one adult *C. elongatus* female with highly developed egg strings. The incubators were installed in a water distribution system where

seawater was supplied and drained from each well continuously. The wells received oxygenated seawater (9.5 ± 0.2 °C) with a water flow of about 20-25 mL/min per incubator. Sampling was performed once a day to check if the lice had shed their exoskeleton in a stereomicroscope until all lice reached the copepodite stage (Stereomicroscope system SZ10, Olympus Corporation). The water temperature, hatch- and molting date were recorded at each examination.

2.3 Infestation of Atlantic salmon with *C. elongatus* and *L. salmonis*

Infection experiments of *C. elongatus* and *L. salmonis* were performed on Atlantic salmon to:

I. Investigate if there is a pattern of where chalimus sea lice attach to the fish.
II. Examine if the sea lice species can be distinguished based on macroscopical features as chalimus larvae.

Nine farmed Atlantic salmons as experimental host fish were provided by the IMR on the 6th of December 2019 in Matre. The fish had a mean length of 33.3 cm, an average weight of 447 g, and were kept in a tank with seawater at 10°C. The salmon were exposed to 1200 copepodites, of which 600 were *C. elongatus* and 600 *L. salmonis*. The sea lice were cultivated and provided by the SLRC at the UoB (Genotype 1). Sampling was conducted and examined on two days. The initial sampling was conducted on four fish (FishID 1-4) 10 days after the infection, and the second sampling was performed on the remaining five fish (FishID 5-9) 15 days after infection.

2.3.1 Attachment sites of chalimi larvae (Part I)

An infection experiment of *C. elongatus* and *L. salmonis* was performed to study if there was a trend of the attachment sites of the sea lice species on Atlantic salmon. The fish's body was divided into eleven areas, including the fins (Figure 5). Each fish (Fish-ID 1-9) was assigned a numbered sheet illustrating the left and right lateral sides of the fish's body surface. The fish was individually euthanized before being placed in a separate white tub filled with fresh seawater, where both lateral sides of the fish were examined for sea lice. The louse found on the fish was given an individual Lice-ID by marking the lice´s position on the fish in the sheet and labeling the lice's microtube with the Lice-ID. The louse was stored in 1.5 mL microtubes filled with 96% ethanol. Subsequently, the sheets with the positions of the lice were categorized into different zones.

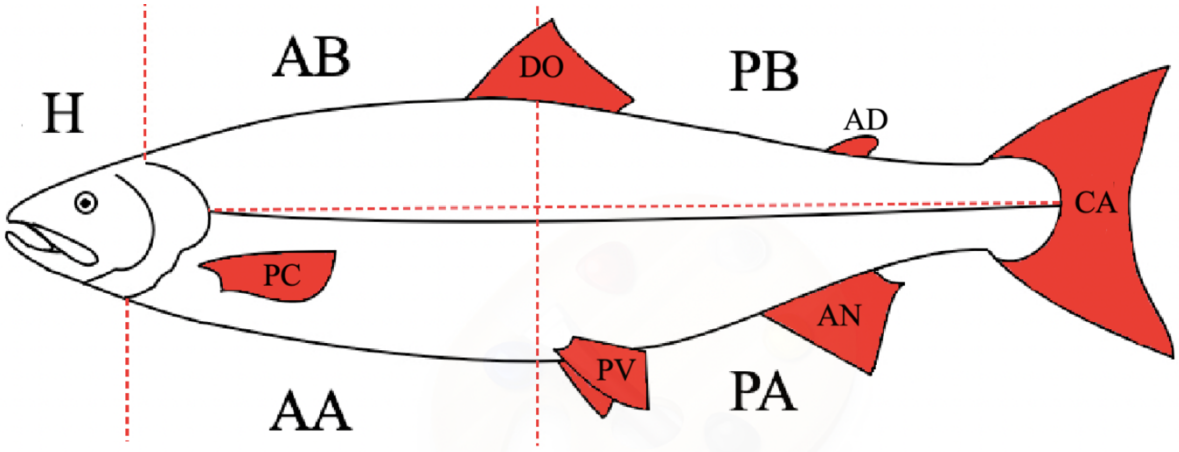

Figure 5. *The Atlantic salmon's body surface, divided into 11 zones: the head (H), anterior abdomen (AA), posterior abdomen (PA), anterior back (AB), and the posterior back (PB). The fins are marked with red color and is separated into dorsal fin (DO), adipose fin (AD), caudal fin (CA), anal fin (AN), pelvic fins (PV), and pectoral fins (PC).*

2.3.2 Macroscopical identification of chalimus larvae (Part II)

The sea lice species *C. elongatus* and *L. salmonis* often co-occurs and it can be challenging to separate the species macroscopically in the sessile stages. However, previous research has reported differences between *C. elongatus* and *L. salmonis* in the chalimus stages. This experiment examined the extent to which it was possible to distinguish and correctly identify *L. salmonis* and *C. elongatus* in the early chalimi stages based on the reported differences. Fish, lice, and sampling days are the same as described in the Atlantic salmon infestation experiment (2.3).

The fish were first euthanized before each louse was photographed on the fish with a camera (Canon EOS 2000D 18-55MM, Japan). Morphological characteristics in the chalimi stages of the two sea lice species were made based on descriptions from previous research of the sea lice (Table 2). Based on the different characteristics, the lice species were categorized as either *C. elongatus* or *L. salmonis* to examine if the characteristics could be used to separate the species. This was performed with eight random lice from each of the nine infected fishes, except for fish number one and two with five and six sea lice, which makes a total of 67 sea lice. The correct identification of the lice was subsequently revealed by a Polymerase Chain Reaction (PCR). A PCR amplifies a specific part of the DNA segment by a repetitive cycle of denaturation, annealing, and extension. The process starts with separating the DNA strands from another (denaturation) using high temperatures. Next, the oligonucleotide primers attach the template as the temperature drops (annealing). When the temperature rises again, the

sequence is copied (elongation). The number of cycles represents the desired amount of copies of the DNA fragment (Kubista et al., 2006).

Table 2. *Morphological differences between C. elongatus and L. salmonis at their respective developmental stage at sampling 1 (S1) and 2 (S2). The sea lice were expected to be chalimus I at the first sampling, and C. elongatus was expected to be chalimus III and L. salmonis chalimus II at the second sampling.*

Morphological differences in chalimus stages of sea lice	
C. elongatus	***L. salmonis***
Chalimus I (S1)	Chalimus I (S1)
- Golden-brown body pigmentation	- Brownish body pigmentation
- Bright red colored eyespots	- Dark red colored eyespots
- Pigmented area around eyespots	- Unpigmented area around eyespots
- The anterior tip of the cephalothorax is sharper than *L. salmonis*	- The anterior tip of the cephalothorax is flatter than *C. elongatus*
- Longer abdomen than *L. salmonis*	- Shorter abdomen than *C. elongatus*
- Slightly smaller than *L. salmonis*	- Slightly larger than *C. elongatus*
Chalimus III (S2)	Chalimus II (S2)
- Smaller than *L. salmonis*	- Larger than *C. elongatus*
- Long, slender frontal filament	- Short, thick frontal filament

2.3.2.1 Multiplex PCR and gel electrophoresis

DNA extraction, multiplex PCR, followed by an agarose gel electrophoresis was performed to obtain inambiguous sea lice species identifications of *C. elongatus,* and *L. salmonis*. DNA was extracted by heating the sea lice with water (a procedure used by the IMR). Each louse was placed in a well on the PCR plate, and the whole lice were covered in 30.0 µL dH_2O. The PCR plate was heated up to 99°C for 10 min. Then, the PCR plate was spun in the centrifuge at 6000 x g for 2 min, and 3.0 µL of the supernatant was used as a template in the multiplex PCR. The PCR multiplex mix was performed with GoTaq Flexi DNA Polymerase (Promega Corporation, USA), according to the manufacturer's standard application protocol. The reaction consisted of 5.00 µL 5x GoTaq Flexi DNA Polymerase, 2.50 µL $MgCl_2$ [25 mM], 4.00 µL dNTPs [25 mM], 4.85 µL dH_2O, 6.00 µL template DNA and 0.63 µL [10 µM] forward and reverse primers in a total volume of 25.00 µL. The primers used were LsF1939, LsR1941, CeF1940, and CeR2948, targeting mitochondrial cytochrome oxidase I (mtCOI) (Table 3) (Mcbeath et al., 2006). PCR conditions used were as follows: activation of the PCR DNA polymerase for 5 min at 95°C, template denaturation for 30 sec at 95°C, primer annealing for 1 min at 55°C, and fragment

elongation for 1 min at 72°C. Step 3–5 was repeated 35 times, followed by a final elongation for 5 min at 72° C (Table 5).

Table 3. *Primers used to distinguish C. elongatus (Ce) from L. salmonis (Ls) with forward (F), and reverse (R) primers, and 5' to 3' sequences.*

Primer name	Primer sequences (5' to 3')
CeF1940	ggcatttcct cgcctgaata
CeR2948	ccaatatacc taaacaccga
LsF1939	gacatagctt tcccccgctta
LsR1941	ggcatttcct cgcctgaata

The multiplex PCR combined two primer pairs used to obtain PCR products with different base pair (bp) sizes for *C. elongatus* (257 bp) and *L. salmonis* (102 bp). The species can be distinguished molecularly by comparing the size of the DNA fragments from the different species on an agarose gel. The contrast in the number of base pairs is large enough for the bands to appear on different areas on the gel, where the hits of *C. elongatus* with 257 bp are located at a higher position than to *L. salmonis* with 102 bp. Gel electrophoresis in 1% Seakem LE agarose (BioNordica, art. nr. L 50004) with GelRed 10.000X in water (VWR, art. nr. 730-2960) was used to visualize products and the size of the PCR products in order to determine the sea lice species. The 1% agarose gel was made by boiling 1.6 g Low Electroendosmosis (LE) agarose (Sigma-Aldrich, USA) and 160.0 mL 0.5 x Tris-acetate EDTA (TAE) buffer (Sigma-Aldrich, USA). 8.0 µL GelRed was added and mixed gently before the agarose gel was poured into the gel-casting container (Sub-Cell® GT Agarose Gel Electrophoresis Systems, Bio-Rad) with a comb to solidify for 10 min. The gel was placed in an electrophoresis chamber, and a 0.5 x TAE buffer was poured over the gel until it covered the agarose gel. The comb leaves wells in the agarose gel, which were loaded with 4.0 µL PCR product and 2 µL loading buffer. 3.5 µL DNA Ladder Mix (MassRuler, Thermo Fisher) was added to each side of the PCR product to determine the fragment size. The agarose gel was run at 110V for one hour (Electrophoresis Power Supply EPS-300, Sweden). When the agarose gel was finished, the gel picture was captured (iBright CL 1000 Invitrogen imaging system, USA). The resulting (true) identities of the chalimi larvae were then compared with the assumed sea lice species based on the macroscopical examination.

2.4 Identification of previous hosts of *C. elongatus*

An attempt was made to identify DNA from the previous host of *C. elongatus* in the gut content of the lice. The cytochrome c oxidase subunit 1 (COI/CO1/COX) gene has been used as a method for species identification through "DNA barcoding" for many different animal species (Hebert, Cywinska, Ball, & DeWaard, 2003; Wootten et al., 1982). The "DNA barcoding" method compares a short fragment of an unknown host's COI gene with genetic material (DNA) from known host species in a quality-assured DNA barcode library to identify the correct animal species (Wilson, Sing, & Jaturas, 2018). *Caligus elongatus* was collected from Atlantic salmon, sea trout, Arctic char, lumpfish, grey gurnard, and garfish caught as bycatch in the salmon surveillance program to examine if the previous host of the lice could be identified by this method.

2.4.1 Molecular taxonomic method

A pilot study was performed to examine if it is possible to molecularly identify the previous host's DNA from the gut contents of *C.* elongatus as the lice grazes on blood and mucus from its host. This method was performed on lice where the previous host was known to test if the method could be used to identify the previous host of *C. elongatus* on lice with an unknown host in the future. The laboratory procedure was tested as a pilot study with ten samples of *C. elongatus* found on grey gurnard, lumpfish and garfish collected from Låva at Boknafjord, Rogaland. An alignment of the COI genes of the host species and *C. elongatus* was made in the software CLC Genomics Workbench (QIAGEN Digital Insights, 2014) to test if the COI gene for *C. elongatus* was sufficiently incompatible from the host species to be amplified by the primers used (Appendix D - Table 1-4D).

For DNA isolation, the sea lice were put on paper to remove as much ethanol (EtOH, 96%) as possible and then cut into six smaller fragments with a sterile carbon steel scalpel blade. The paper was replaced, and the scalpel was wiped with ethanol between each sample. The fragments of the louse were placed in 1.5 mL microtubes. DNA extraction was conducted with the DNeasy® Blood and Tissue kit (Qiagen, Germany) (QIAGEN, 2020) following the manufacturer's animal tissue purification protocol (DNeasy 96). The instruments used are listed in Appendix A–Table 5A. DNA sample's concentration and purity were measured by analyzing the absorbance (A) of wavelengths at 260/280 nm (A_{260}/A_{280}) and 260/230 nm (A_{260}/A_{230}) nm in a spectrophotometer (NanoDrop® ND – 1000, USA) (Appendix C). The DNA extracts were stored in a 4°C refrigerator.

A PCR amplified the COI target, and the reactions were carried out using a GoTaq Flexi DNA Polymerase kit (Promega Corporation, 2012) according to the standard application described in the protocol. The master mix contained 2.40 µL 5x GoTaq Flexi Buffer, 1.00 µL $MgCl_2$ [25 mM], 1.92 µL dNTPs [25 mM], 0.07 µL GoTaq Flexi DNA Polymerase, 3.17 µL dH_2O, 2.00 µL DNA template, and 1.44 µL [10 µM] primer pair combination containing the M13 sequence in a total volume of 12.00 µL (Table 4) (Ivanova, Zemlak, Hanner, & Hebert, 2007; Mateos-Rivera et al., 2020). PCR conditions for the GoTaq PCR amplification were as follows: activation of the DNA polymerase for 5 min at 95°C, repetition of 35 cycles of template denaturation for 30 sec at 95° C, primer annealing for 1 min at 52°C, and fragment elongation for 1 min at 72°C, followed by a final elongation for 5 min at 72°C, and then 4°C until the sample was collected (Table 5) (GeneAmp PCR system 9700, Applied Biosystems).

Table 4. *COI primer set used to detect the COI gene of fishes found in the North Sea. Forward (F), and reverse (R) primers and 5' to 3' sequence. Additional M13 sequencing primers are highlighted in red (F) and blue (R).*

Primer name	Primer sequences (5' to 3')
COI-2-LepF1_t1	tgtaaaacgacggccagt attcaaccaa tcataaagat attgg
COI-2-VF1_t1	tgtaaaacgacggccagt tctcaaccaa ccacaaagac attgg
COI-2-VF1d_t1	tgtaaaacgacggccagt tctcaaccaa ccacaargay atygg
COI-2-VF1i_t1	tgtaaaacgacggccagt tctcaaccaa ccaiaaigai atigg
COI-2-LepR1_t1	caggaaacagctatgac taaacttctg gatgtccaaa aaatca
COI-2-VR1_t1	caggaaacagctatgac tagacttctg ggtggccaaa gaatca
COI-2-VR1d_t1	caggaaacagctatgac tagacttctg ggtggccraa raayca
COI-2-VR1i_t1	caggaaacagctatgac tagacttctg ggtgicciaa iaaica

Clean-up of the PCR products was performed by mixing 5.0 µL PCR product with 2.0 µL ExoSap-IT PCR product (art. nr. US77702, VWR) followed by a PCR program of incubation at 37°C for 15 min to degrade the remaining primers and nucleotides and 80°C for 15 min to inactivate the reagent, followed by 4°C until the sample was collected (Table 5).

M13 primers targeting the binding seat of the DNA targeting primers were used as sequencing primers. 1.0 µL Big Dye Buffer, 1.0 µL Big Dye, 4.0 µL H_2O, and 3.0 µL purified PCR product were mixed in microtubes in a total of 9.0 µL. Each of the ten samples was split into 20 vials. 1.0 µL M13 forward primer was added to ten of the vials, and 1.0 µL M13 reverse primer was added to the remaining ten vials, which gives a total of 10.0 µL in each sample. The reactions were run for BigDye PCR sequencing with the following conditions: initial

denaturation at 95°C for 5 min, a repetition of 28 cycles at 95°C for 10 sec, 50°C for 5 sec, and 60°C for 4 min, and then 4°C until the sample was collected (Table 5).

Finally, sequencing the PCR products was carried out by the sequencing facility (http://www.seqlab.uib.no) (3730xl DNA Analyzer, Applied Biosystems). The sequence was trimmed and manually controlled before the samples were run in the software Nucleotide Basic Local Alignment Search Tool (BLASTn) in the National Center for Biotechnology Information (NCBI) to examine if the sequences could be identified.

Table 5. *Three PCR settings with the temperature (°C), time (minutes/ seconds), and the number of cycles at each PCR step. All except Exo-Sap-IT have repeated cycles (→) of different steps. Infinity symbol (∞) refers to a setting where the sample remains at a specific temperature until the samples are removed from the machine. GoTaq DNA Polymerase PCR conditions to find the previous host of C. elongatus used an annealing temperature (*) of 52°C instead of 55°C.*

Thermal cycler conditions						
GoTaq® DNA Polymerase-Mediated PCR amplification						
Step	1	2	3	4	5	6
Temp (°C)	95	95	55*	72	72	4
Time	5 min	30 sec	1 min	1 min	5 min	∞
Cycle		→35				
ExoSap-IT™ PCR Product Cleanup						
	Step		1	2	3	
	Temp (°C)		37	80	4	
	Time (min)		15	15	∞	
BigDye® Terminator v3.1 PCR program						
Step	1	2	3	4	5	
Temp (°C)	95	95	50	60	4	
Time	5 min	10 sec	5 sec	4 min	∞	
Cycle		→28				

2.4.2 PCR gradient to reduce *C. elongatus* amplification

A PCR gradient was attempted to investigate if it is possible to reduce the amplification of *C. elongatus* in order to detect the fish's COI gene. A PCR gradient of 50, 52, 54, 56, 58, and 60°C was tested on four *C. elongatus* samples as a test to confirm if an increased annealing temperature reduced the amplification of *C. elongatus* or not. Prior hosts of the samples were lumpfish (3, 4), and garfish (8, 9). Sample 3 had a short sequence, sample 4 had a clear sequence

(control), sample 8 had a lot of baseline noise, and sample 9 had a lot of double sequences (Appendix G - Figure 1-4G). The samples used in the PCR gradient (3, 4, 8, 9) were run in the ExoSap-IT PCR product clean-up followed by a BigDye PCR, both performed as described in the manufacturer's protocols (2.4.1) (Table 5). Finally, sequencing was performed at the sequencing facility (UoB). A 1% medium agarose gel was made using the same procedure as described previously (2.4.1) but with a 0.8 g LE buffer, 80.0 mL 0.5 TAE buffer, and a 3.5 µL GelRed Nucleic Acid Gel Stain. The wells were filled with 4.0 µL PCR product and 2.0 µL 5x Green GoTaq Flexi Buffer. The agarose gel was run at 80V for 50 min before it was photographed (iBright CL 1000 Invitrogen imaging system, USA).

An overlapped PCR gradient at 58, 60, 62, 64, 66, 68°C was created to remove the unwanted amplification of *C. elongatus* as results from the first PCR gradient (50-60°C) showed strong bands at all temperatures. In addition, the sequences received from the sequencing facility from the first PCR gradient could not be identified in BLASTn, which means there still are problems with unwanted amplification of *C. elongatus* DNA. (Figure 17). The second PCR gradient was performed as the first one, but with different annealing temperatures.

2.4.3 Saithe experiments

Two laboratory experiments of adult *C. elongatus* was performed to examine how the DNA in the intestinal content of saithe develops in the lice after:

I. The lice have re-infected farmed Atlantic salmon and stayed on the new host for different time intervals.
II. The lice have been unattached from the host to starve in water for different time intervals.

Saithe were collected by fishing with a fishing rod close to a fish farm at Austevoll, Hordaland. Sampling was performed after time intervals of 1, 3, and 22 hours for both experiments. DNA extraction, PCR, and gel electrophoresis were performed to visualize the results from the experiments. The experiments were performed at the IMR station at Austevoll.

Transmission of *C. elongatus* from saithe to salmon

An experiment was performed to investigate if it is possible to detect saithe DNA from the gut contents of *C. elongatus* initially found on saithe and subsequently transferred to Atlantic salmon for 1, 3, and 22 hours. *Caligus elongatus* was collected from saithe fished with a fishing rod at Austevoll, Hordaland, from the 30th to the 31st of October 2019. The saithe were visually examined for adult *C. elongatus* in the field, the lice were removed from the fish and placed in a labeled 50 mL corning centrifuge tube filled with fresh seawater. Eighteen adult lice were collected from seven fishes. The IMR supplied six farmed Atlantic salmons smolts with an average weight of 63 g that were placed in a separate fish tank in the wet lab.

The salmon were individually anesthetized in a bath of Tricaine-S MS 222 (Tricaine Methanesulfonate) (Syndel, USA) in a tub filled with seawater. The anesthetized fish was then transferred into a small tub with fresh seawater, where *C. elongatus* from the saithe was added to infect the salmon. The number of lice that settled on the fish was recorded and the salmon was carefully released back in the original fish tank. The sea lice were removed from the salmon and reserved in microtubes filled with 96% ethanol after 1, 3, and 22 hours for further molecular analyses. The fish was euthanized with an overdose of Tricaine-S MS 222 when the experiment was finished.

The molecular procedure with DNA extraction, PCR, and gel electrophoresis was the same for the transmission and starvation experiments. Both saithe experiments were performed as described in the pilot study to identify previous hosts of *C e.ongatus* (2.4.1), except that the primers were replaced with a specific primer pair for saithe (Table 6) (Nilssen et al., 2019). The specific saithe primer was used to avoid detecting *C. elongatus,* the dominating DNA in the sample. In addition, four negative controls with DNA of Atlantic salmon, *C. elongatus*, *L. salmonis*, H_2O, and four positive control samples of DNA from saithe (received by the IMR) were added to the agarose gel. PCR conditions used were as follows: activation of the PCR DNA polymerase for 5 min at 95°C, template denaturation for 30 sec at 95°C, primer annealing for 1 min at 55°C, and fragment elongation for 1 min at 72°C. Step 3–5 was repeated 35 times, followed by a final elongation for 5 min at 72° C, then 4°C until the sample was collected (Table 5). A large 1% agarose gel was made, and gel electrophoresis was run for one hour at 120V. An iBright CL 1000 Invitrogen captured a gel picture when the agarose gel was finished (Thermo Fisher Scientific, USA).

Table 6. *Specific primer pair used to detect saithe in C. elongatus. Primer name including forward (F), and reverse (R) primers, and 5' to 3' sequence.*

Primer name	Primer sequences (5' to 3')
Saithe-F	gaatcccaat aattttaata gcct
Saithe-R	tcgattgctt agtcatcgag a

Starvation of *C. elongatus* from saithe

An experiment was performed to investigate the time interval of which it is possible to identify DNA from saithe in the gut contents of *C. elongatus* starved in water for 1, 3, and 22 hours. A total of ten lice was collected from six saithe fished with a fishing rod near a fish farm at the IMR station at Austevoll, Hordaland, 16th and 17th of November 2019. The lice were carefully put in 50 mL corning centrifuge tubes filled with fresh seawater. The exact time when the lice were collected from the fish was recorded and the tubes were marked. The lice were kept alive for 1, 3, and 22 hours before the lice were preserved in 96% ethanol in labeled, 1.5 mL microtubes. DNA extraction, PCR, and gel electrophoresis was performed as described in the transmission experiment (2.4.3).

2.5 Statistical analysis

Data analyses were performed using RStudio version 1.2.5033 (RStudio Team, 2019) and Statistica™ version 13 (TIBCO Software Inc, 2017). Additional packages for RStudio for the generalized linear models (glm), violin plot, boxplot, and bar charts: Tidyverse (H Wickham, 2017), extrafont (Chang, 2014), RcolorBrewer (Neuwirth, 2014) readxl (H Wickham & Bryan, 2019), dplyr (Hadley Wickham, François, Henry, & Müller, 2020) and ggpmisc (Aphalo, 2019). Extra packages to create the map in Figure 3: rgdal (Bivand, Keitt, & Rowlingson, 2019), ggmap (Kahle & Wickham, 2013), and ggrepel (Slowikowski, 2019).

Analyses performed in Statistica: T-tests were performed to examine if there was a significant difference in lice length and the number of eggs based on the different regions, and to test if it was significant difference between the duration of nauplius I and II stage of *C. elongatus*. Furthermore, a Mann-Whitney U test was performed to investigate whether there is a difference in the number of eggs based on the origin of the lice's host, and if there is a difference in the number of eggs for farmed and wild origin in southern Norway. Three different Kruskal-Wallis tests were performed. The first test examined if there was a significant regional difference in the number of eggs for lice from wild fish. The second test investigated if there was a significant difference in the number of eggs for lice with different host species. The last

test examined if there was a significant difference in the lice length of lice from different host species. These tests were followed by a Post Hoc multiple comparisons test that defined where the variation was. A Pearson's coefficient correlation was calculated in order to examine if there was a significant correlation between the egg string length and the number of eggs for different host, origins, and geographical regions. The RStudio script and Statistica outputs are listed in Appendix H, and the dataset used is found in Appendix I. All coding was generated by the author (SMHR).

3 Results

3.1 Investigation of *C. elongatus* fecundity

Lice from wild vs. farmed fish

The egg string length and the total number of eggs in them were compared between lice from 'farmed' and 'wild' origin (Figure 6, a). A Pearson´s correlation coefficient proved a significant correlation between the number of eggs and the egg string length and explained 53% (r^2=0.53) and 17% (r^2=0.17) of the data variation in the lice from farmed and wild fish, respectively. The regression line for lice with farmed origin is steeper than lice from wild origin and lice from wild fish had a higher incidence of outliers than lice from farmed origin. Most egg string lengths from farmed *C. elongatus* ranged between 3-4 mm (mean 3.5 mm) and contained 75-125 eggs. The majority of the lice from wild origin had egg string lengths ranging from 5-7 mm (mean 5.3 mm) and contained 80-140 eggs. A Pearson's correlation coefficient found a significant positive correlation between the egg string length and the number of eggs for lice from wild (r^2=0.22, N=179, p<0.001) and farmed fish (r^2=0.53, N=81, p<0.001) in southern Norway. Lice from northern (r^2=0.14, N=22, p=0.08) and central Norway (N=6) did not prove a significant correlation between the egg string length and the number of eggs.

There was no correlation between the body size and the number of eggs from wild and farmed origin (Figure 6 b). A t-test revealed no significant variance between lice length and the number of eggs for lice from northern (T_{21}=1.133, p=0.271) and central (T_5 =0.588, p=0.588) Norway. There is significant variation between the sea lice size and the number of eggs from lice from southern Norway, both wild (T_{178} =2.034, p=0.043), and farmed (T_{80} =2.175, p=0.033) origin, with an r^2 value of 0.023 and 0.057, respectively. A t-test revealed a significant difference in the lice length between farmed and wild origin (T_{287}=6.455, p=0.000). The average body length of lice from farmed and wild origin was 5.4 mm (SD=0.52) and 5.8 mm (SD=0.45), respectively (Appendix H).

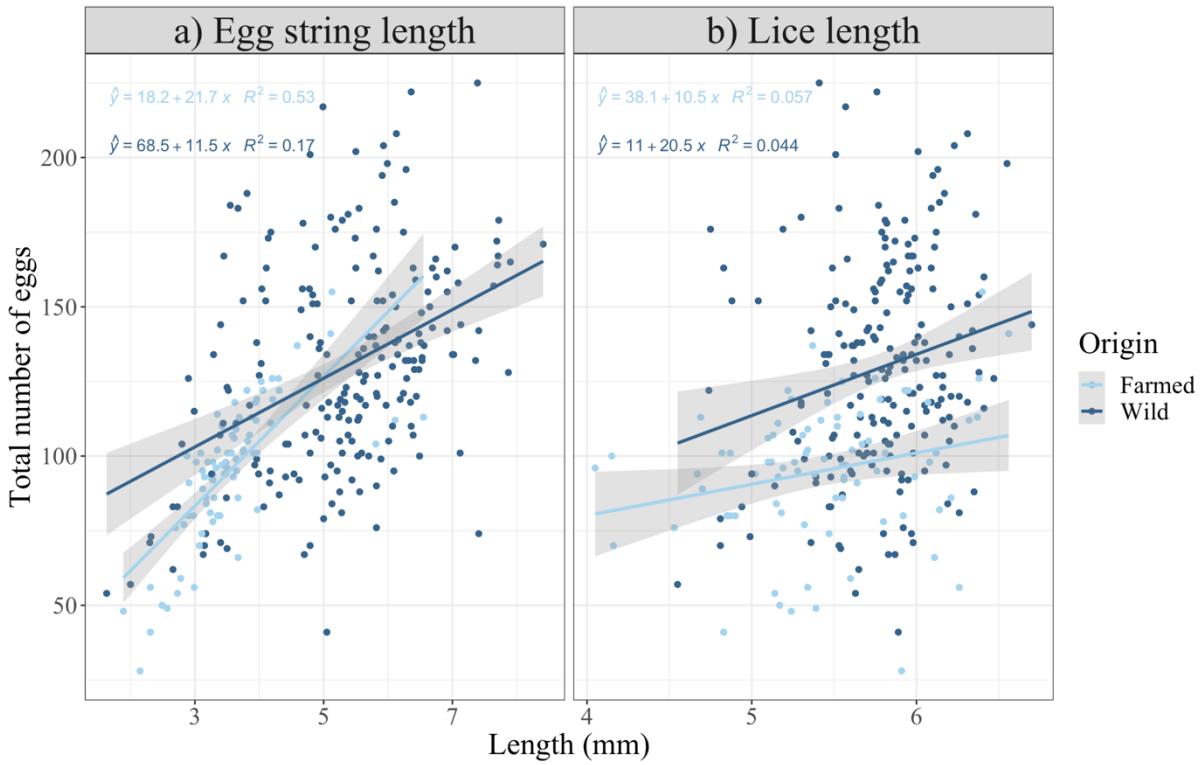

Figure 6. *Linear regression models of the number of C. elongatus eggs compared to a) the egg string length (mm) and b) the total length of the louse (mm). The dots represent lice with wild (dark blue) and farmed (light blue) origins. The 95% confidence interval of the model is the shaded area around each of the regression lines. The upper left corner shows the linear regression equation (Y=a+bx) and the coefficient of determination (R^2).*

There was no difference between the number of eggs or the length of the left and right egg strings for lice from farmed and wild origin. The average number of eggs per mm egg string provides information on the size of the eggs. Lice originating from farmed and wild fish had an average of 12.7 and 13.4 eggs per mm egg string.

Table 7. *An overview of the number of eggs and the length (mm) of the egg strings (Left / Right) of C. elongatus of farmed and wild origin. x (y, n=z), x=mean, y=SD, z=number of lice.*

Number of eggs and egg string length				
Parameters	**Farmed**		**Wild**	
Egg string	Left	Right	Left	Right
Number of eggs	48 (12, n=81)	47 (12.14, n=81)	65 (18.19, n=208)	65 (18.33 n=208)
Length of egg string (mm)	3.55 (0.82, n=81)	3.54 (0.76, n=81)	5.35 (1.32 n=208)	5.30 (1.31 n=208)
Nr. eggs pr. mm egg string	13.4 (2.15, n=81)		12.7 (2.82 n=208)	

There was a highly significant difference between the number of *C. elongatus* eggs from farmed Atlantic salmon and wild fish (Mann-Whitney $Z_{(N\ wild=207,\ N\ farmed=81)}=7.67$, $p<0.001$) (Figure 7). Lice from farmed origin have most eggs ranging between 82 to 110 eggs. The data's interquartile range (IQR) (Q_3-Q_1) was 28 eggs, and the median was 98 eggs per lice. There are two outliers from farmed origin with 26 and 152 eggs. Lice from wild origin had most eggs ranging from 104 to 154 eggs with an IQR of 50 eggs and a median of 130 eggs per sea lice. Lice from wild fish show more data variation than lice from farmed origin. Calculations of the fecundity of *C. elongatus* are based on data from Appendix I.

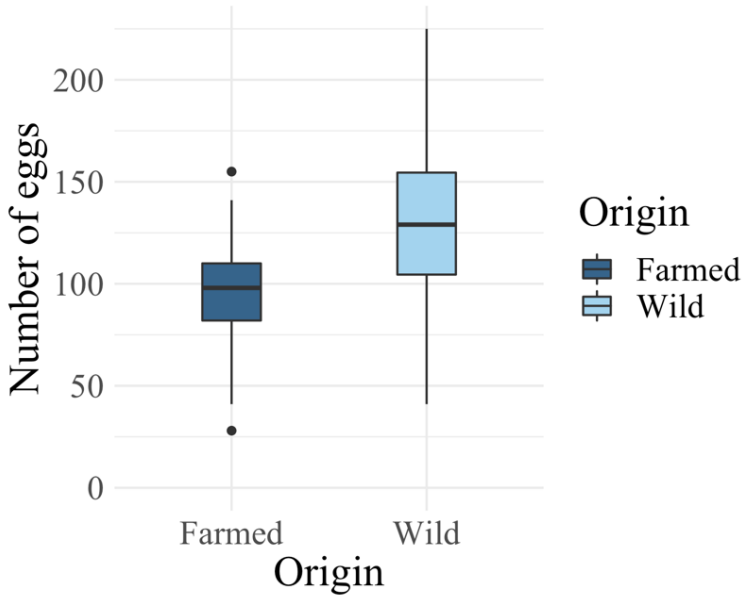

***Figure** 7. Boxplot of the number of eggs from farmed (dark blue) and wild (light blue) origin. The black dots represent outliers. The number observations in parenthesis: farmed (81) and wild (207).*

Body length and the number of eggs of lice from different hosts

The body length and the number of eggs in the egg strings of *C. elongatus* were measured for lice from five different host species: Atlantic salmon, Arctic char, lumpfish, sea trout, and garfish (Figure 8). The number of *C. elongatus* eggs from the majority of the different host species ranged between 80-180 eggs. A Kruskal Wallis test proved there was a significant difference between the number of eggs and the lice host (KW, $H_{(4,\ n=285)} = 5.122$, $p=0{,}275$). A Post Hoc multiple comparisons test revealed a significant difference in the number of eggs between lice from Atlantic salmon and the remaining host species. There was a significant difference in the number of eggs between garfish and Atlantic char, and garfish and lumpfish.

Lice found on Arctic char had, on average, the most eggs, followed by sea trout, lumpfish, garfish, and Atlantic salmon. However, it is important to notice that there were limited data from Arctic char, lumpfish, and sea trout. There was a bimodal distribution for Arctic char with increased frequencies at 110 and 180 eggs, whereas the latter had the highest frequency. Arctic char had a left-skewed distribution with a median of 176 eggs and an IQR of 30 eggs. Sea trout showed a bimodal distribution with modes of 95 and 170 eggs, respectively, and a left-skewed distribution. Sea trout shows the largest IQR of the host examined with 82 eggs. Lice from lumpfish showed great variance in the number of eggs, evenly distributed from 57 to 222 eggs, an IQR of 56 eggs, and a median of 151 eggs. Both farmed Atlantic salmon and garfish had the least eggs with a normally distributed plot and an IQR of 29 and 36 eggs, respectively. The shape of the violin plot indicates that the two latter species are highly concentrated around the median value of 98 and 123 eggs. They had more observations compared to the other species examined.

A Kruskal Wallis test proved no significant difference between the lice length and the host (KW, $H_{(4, n=285)} = 5.122$, p=0,275). The average body length of lice from the different host species varies by less than 0.5 mm. Lice from sea trout have a slightly right-skewed length distribution with an IQR of 0.57 mm. Lice from garfish and lumpfish present very similar plots, where both are normally distributed with mean lengths of about 5.8 mm. The IQR of garfish (0.35) is slightly lower than lumpfish (0.50). The plot for lumpfish is interfered with two outliers at around 4.6 mm. Lice from the distribution of lice from Arctic char are right-skewed and have the highest density with a body length of around 6.3 mm and a slightly less dense area at about 5.3 mm. The IQR of lice from Arctic char is 0.46 mm, and the highest mean value of all hosts with 6.2 mm. Lice from Atlantic salmon has an even distribution and an IQR of 0.62 mm. Most observations range from 5.1-5.8 mm (Figure 8).

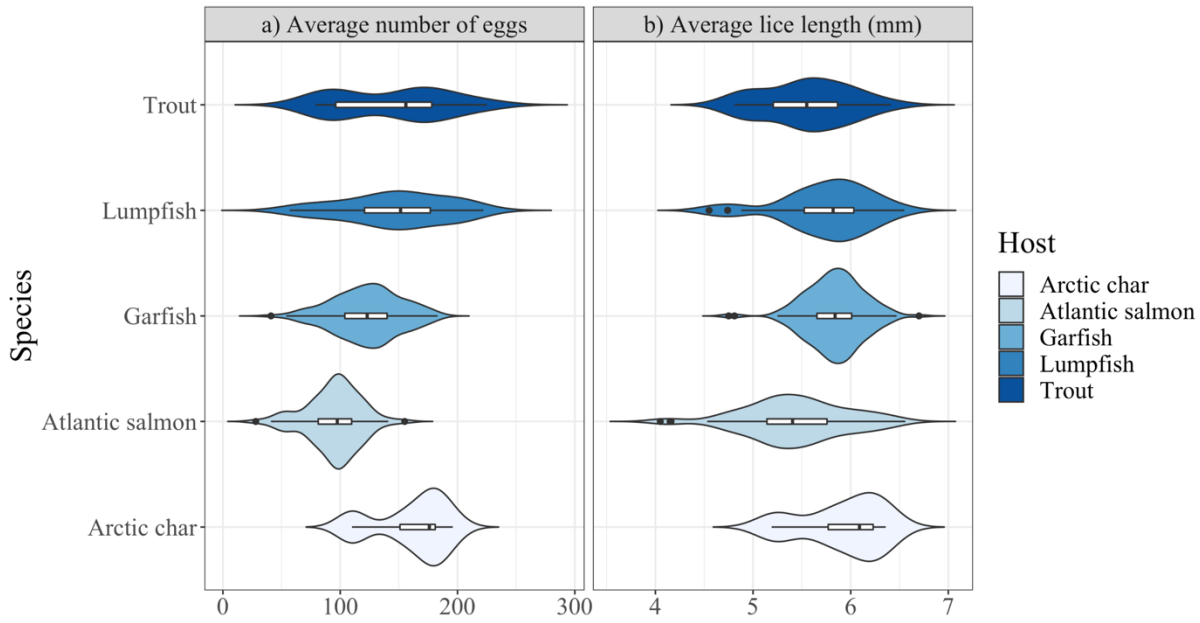

Figure 8. *A violin plot of* ***a)*** *the average number of C. elongatus eggs and* ***b)*** *the average lice length (mm) compared to the host species of C. elongatus. The black dots represent outliers. The number of observations in parenthesis: Arctic char (9), lumpfish (28), sea trout (22), garfish (143), and Atlantic salmon (82).*

The numbers of eggs and geographical regions

There was a highly significant difference in *C. elongatus* eggs from farmed Atlantic salmon and wild fish ($p<0.0001$) as revealed in the Mann-Whitney U test. A Kruskal-Wallis test proved the variation between the number of eggs and the regions (KW, $H_{(2,\ n=\ 207)} = 13{,}5$ $p=0.0089$). A Post Hoc multiple comparison test confirmed a significant difference between lice from southern Norway compared to the central- and northern parts of Norway. A Mann-Whitney U test revealed a highly significant difference in the number of *C. elongatus* eggs from farmed Atlantic salmon and wild fish, both from southern Norway (MW, $Z_{(N\ wild=179,N\ farmed=81)}=7.08$, $p<0.001$).

The average number of *C. elongatus* eggs at different sites along the Norwegian coast shows that Altafjorden and Reisafjorden in the north and Nordfjorden in the south had the highest average of *C. elongatus* eggs, with an average of 160 eggs per lice (Figure 9). The number of materials from all locations except Boknafjorden and Låva is severely limited.

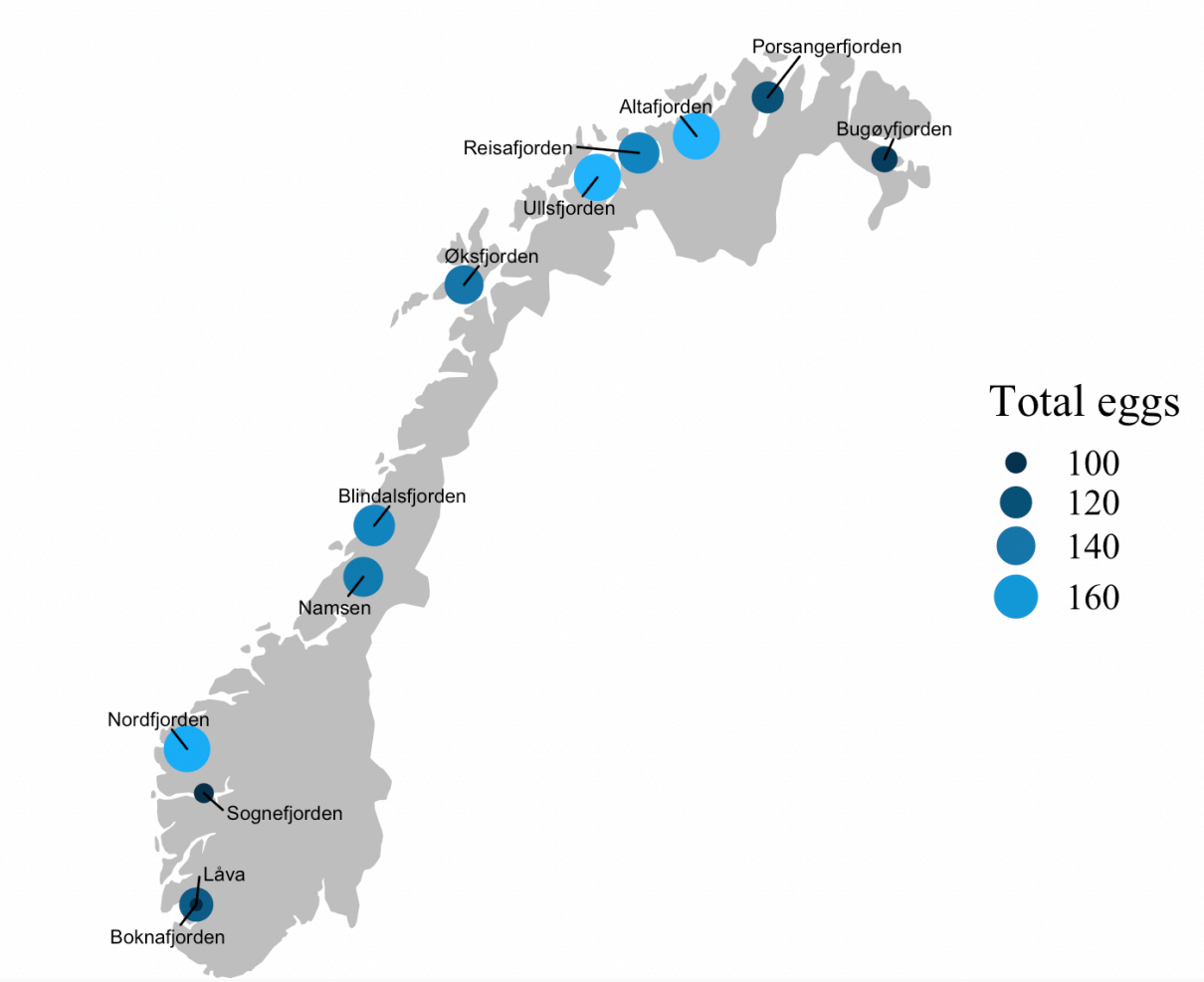

Figure 9. *Bubble map of Norway with locations of the sampling sites. The average number of eggs (100-160) is based on the host's location, and the color represents high (light blue) to low (dark blue) average numbers of eggs where the dots are small for low and larger for higher numbers of eggs. The number of observations in parenthesis: Låva (81), Sognefjorden (5), Bugøyfjorden (2), Porsangerfjorden (2), Boknafjorden (166), Øksfjorden (1), Namsenfjorden (1), Reisafjorden (5), Blindalsfjorden (5), Nordfjorden (8), Altafjorden (2) and Ullsfjorden (9).*

3.1.1 Duration of *C. elongatus* nauplius stages

The duration of *C. elongatus* first two nauplius stages was studied under laboratory conditions at a temperature of 9.5°C. The DPH until the lice were molted into a nauplius II larvae (N1) was used to estimate the duration of the nauplius I stage (Table 8 and Figure 10). The days from newly molted nauplius II larvae until the lice molted into copepodites (N2) was used to estimate the duration of the nauplius II stage. The average duration of the nauplius stages of *C. elongatus* was 4.03 days. The average duration of the nauplius I and II stage was estimated to be 1.64 and 2.39 days, respectively. The difference in the duration of the nauplius stages proved to be significant ($p<0.05$).

Table 8. *The duration (days) of C. elongatus nauplius stages (I, II). x (y, n=z), x=mean, y=SD, z=number of lice.*

Developmental stage	Duration (days)
Nauplius I	1.64 (0.83, n=28)
Nauplius II	2.39 (0.74, n=28)
Nauplius I+II	4.03 (0.64, n=28)

Four lice escaped from the incubators during the experiment, which is not included in the data. The majority (60%) of the lice were molted into nauplius II larvae after one day as a nauplius I larvae, 24% at two days, 12% at three days, and 4% at four days. Half of the lice examined (54%) were molted into a copepodite after three days as a nauplius II larvae, 32% spent two days, and 14% spent one day. One louse (Nr. 28) stands out as it was a nauplius I larvae for four days until it molted into a nauplius II larvae, compared to the average of 1.64 days.

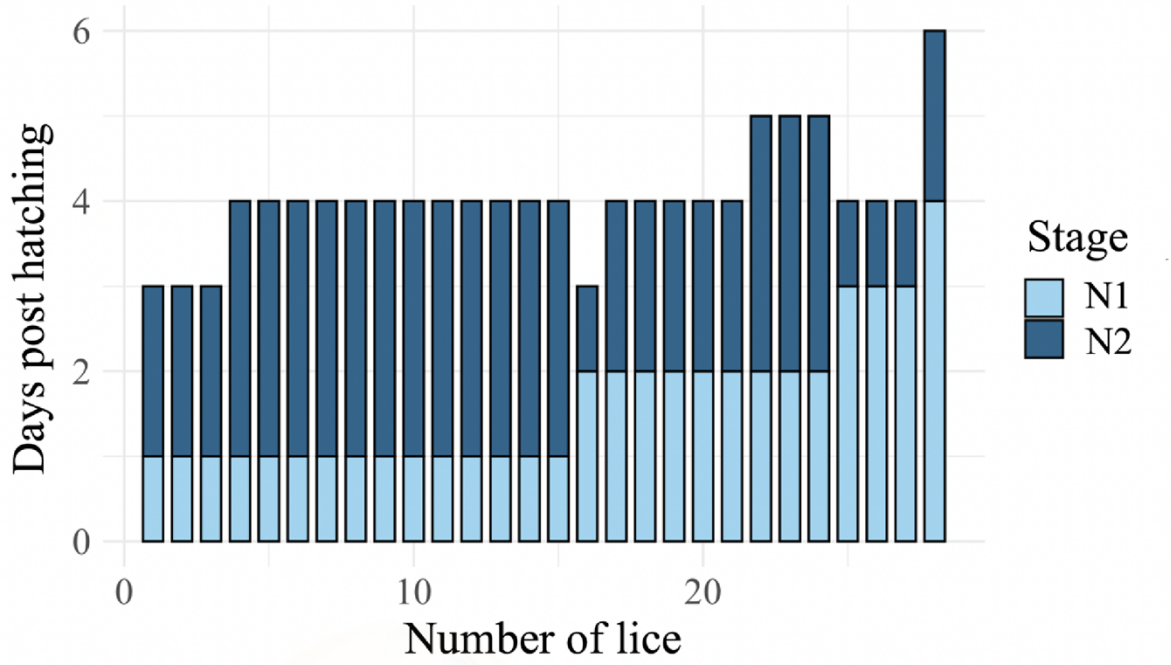

Figure 10. *Duration of the nauplius stages of the offspring from 28 adult, gravid C. elongatus lice at 9.5°C. The bar chart illustrates the DPH until the nauplius I larvae were molted into nauplius II (N1, light blue) and the days from newly nauplius II larvae until molting to copepodites (N2, dark blue).*

3.2 Infestation of Atlantic salmon with *C. elongatus* and *L. salmonis*

3.2.1 Attachment sites of chalimus larvae

Sampling 1

The fish was examined at two sampling dates to investigate whether the sea lice prefer to attach to specific zones on the body surface of the Atlantic salmon (Figure 11). The first sampling consisted of four fishes (Fish-ID-1-4) and had a total of 48 sea lice attached to them. All sea lice were expected to be chalimus I larvae 10 days post-infection (DPI). They were mainly found on the dorsal fin (39%), followed by the posterior back (20%). The sea lice preferred to attach to the fins (61%) rather than the body (39%). Excluding the fish's head and fins, most of the lice attached to the posterior end (82%) compared to the anterior end (18%), and they preferred the dorsal side of the fish (66%) over the ventral side of the fish (34%). All the sea lice attachment sites are listed in Appendix E.

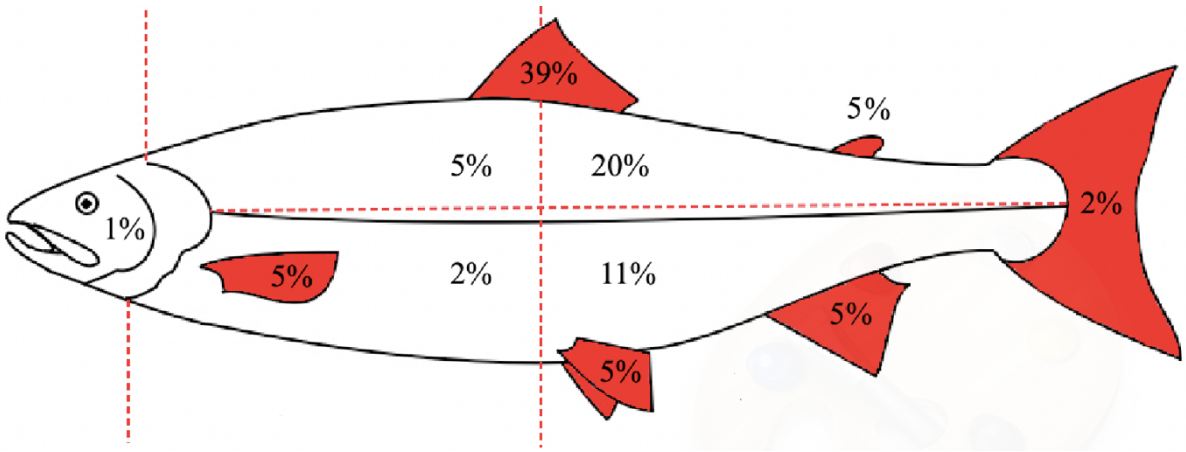

Figure 11. *Atlantic salmon body surface divided into zones and the abundance (%) of sea lice in each area from the first sampling.*

Sampling 2

The second sampling consisted of five fishes (Fish-ID-5-9) and had a total of 88 sea lice attached to the fishes. The lice were expected to be chalimus II (*L. salmonis*) and III (*C. elongaus*) larvae at the second sampling performed 15 dpi. The attachment sites was similar to sampling 1, with an increased sea lice abundance in the fin areas (Figure 12). Most of the lice settled on the fish's fins (74%), where the dorsal fin was favored (54%). The majority of the lice attached to the dorsal side of the fish (83%) over the ventral side (17%) and would rather

attach to the posterior end (87%) of the fish than the anterior end of the fish (13%) with the fins and head excluded.

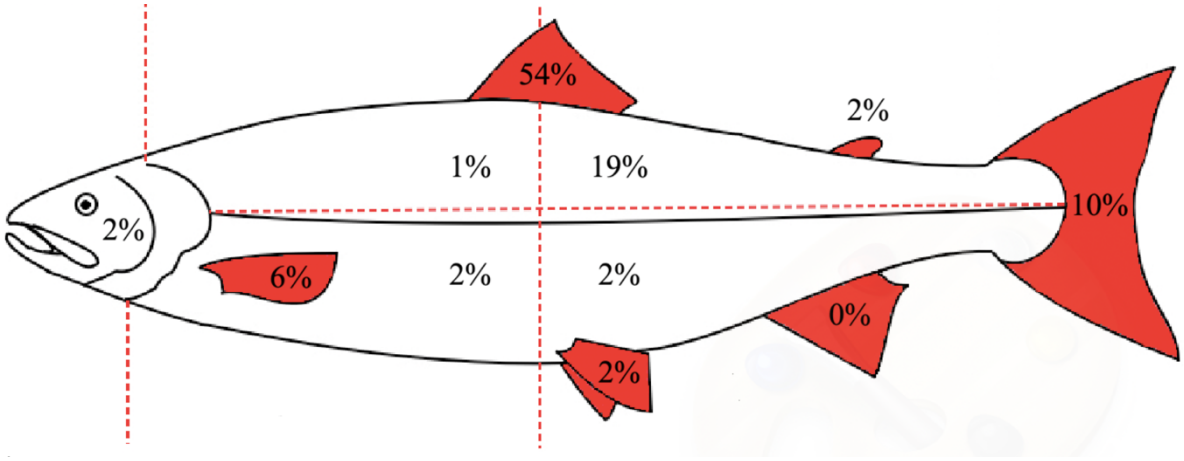

Figure 12. *Atlantic salmon body surface divided into zones and the abundance (%) of sea lice in each area from the second sampling.*

A total of 136 sea lice attached to the nine Atlantic salmon in the experiment. The mean abundance of sea lice from both sampling days (sampling 1, 2) had a generally similar attachment pattern where the lice predominantly favored the dorsal fin (7,7), followed by the posterior back (2,4) (Figure 13). Two zones stood out with a higher abundance of lice in the second sampling compared to the first, both posterior abdomen (0,2) and posterior back (2,4). Sampling 2 had a higher abundance of lice than sampling 1 in all zones except the anal fin and the head. Less than 2 % of the sea lice preferred to attach to the fishes head.

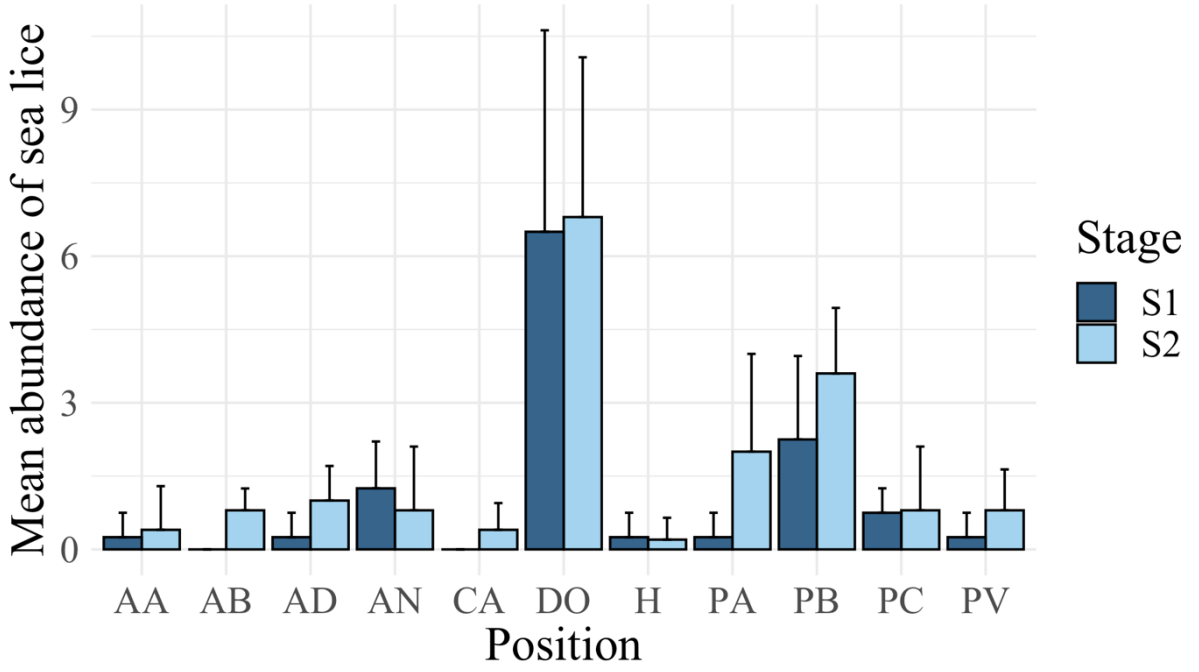

Figure 13. *Bar chart of the abundance of sea lice (L. salmonis and C. elongatus) from the first sampling day (S1, light blue) and the second sampling day (S2, dark blue) (Mean ± SE). Their positions on an Atlantic salmon; anterior abdomen (AA), anterior back (AB), adipose fin (AD), anal fin (AN), caudal fin (CA), dorsal fin (DO), head (H), posterior abdomen (PA), posterior back (PB), pectoral fins (PC) and pelvic fins (PV).*

3.2.2 Macroscopical identification of chalimus larvae

An overview of morphological characteristics was made to help separate *C. elongatus* and *L. salmonis* macroscopically in the chalimus stages. The sea lice species were first tentatively recorded for each chalimus based on a priori listed characteristics. The true species were later identified by molecular identification (control). The vast majority of the assumptions were based on the body pigmentation of the lice, as it was suspected that *C. elongatus* had a bright golden-brown color compared to *L. salmonis* darker brown body pigmentation. Several of the characteristics proved not possible to observe macroscopically. The sharp tip on the anterior part of the cephalothorax was not observed. There were a few observations of a slightly longer abdomen, but no sea lice was identified based on only this feature. The difference in body length of chalimi larvae was too small to determine the sea lice species. Only 0.4 mm separates the species from each other as chalimus I larvae, and 1.2 mm separates *C. elongatus* chalimus III larvae from *L. salmonis* chalimus II larvae (Piasecki, 1996; Pike & Wadsworth, 1999; Schram, 1993). The thickness and the length of the frontal filament were not possible to detect macroscopically and could not help separating the sea lice species. *Lepeophtheirus salmonis* should, in contrast to *C. elongatus* have an unpigmented area around the eyespot. However, the

feature was not visually clear enough to distinguish the species from each other. It appeared that both species had unpigmented areas around the eyes. The eye color helped strengthen the suspicion of sea lice species as *L. salmonis* appeared to have a deeper red, almost black color in contrast to *C. elongatus* brighter red color. Out of the 67 samples studied in the Atlantic salmon infection experiment, 19% were assumed to be *C. elongatus,* and the remaining 81% *L. salmonis* (Appendix B).

The agarose gel image shows that there were bands on 55 wells (82%) out of 67 samples investigated, the remaining 12 wells (18%) were obscure or did not exist on the agarose gel. Lice from the first sampling (FishID1-4) had more vague bands than lice from the second sampling (FishID5-9), and 11 out of the 12 obscure bands originated from the first sampling (Figure 15). *Caligus elongatus* has 257 base pairs (bp), and the band on the agarose gel appears at a higher position on the agarose gel than *L. salmonis* with 102 bp. The difference in numbers of bp separates the species from each other. The PCR result shows that only one louse was confirmed to be *C. elongatus* (1%), it is shown in the agarose gel with a higher position than the other bands on the gel (Figure 14). Fifty-four lice were identified as *L. salmonis* (81%), and the remaining 12 were not detectable on the agarose gel picture (18%). 1,8 % of the successfully identified louse were *C. elongatus* (infection rate = 0.018, $Infection\ rate = \frac{n_{C.elongatus} + n_{L.salmonis}}{n_{C.elongatus}}$).

Figure 14. *Picture of a chalimus C. elongatus louse attached on an Atlantic salmon.*

The only lice correctly identified as *C. elongatus* had a yellowish-light brown color, with slightly reddish-colored eyes. The abdomen was somewhat longer, about half the length of the cephalothorax (Figure 14).

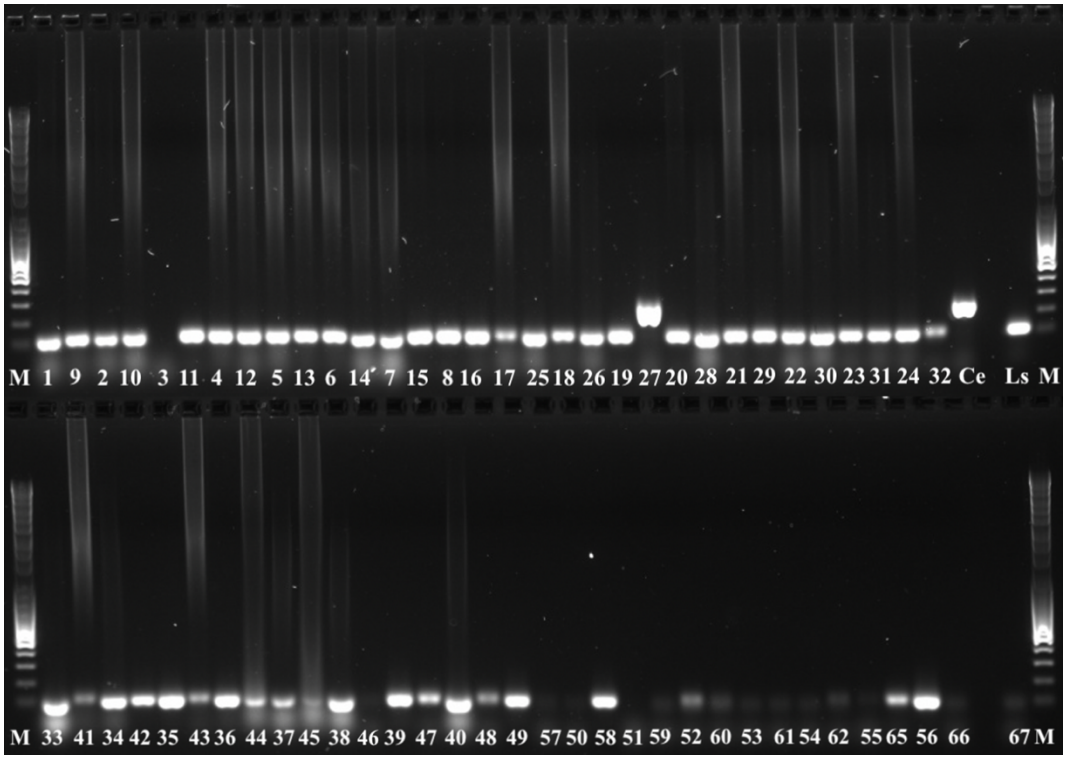

Figure 15. *Agarose gel electrophoresis result of 67 samples of sea lice (C. elongatus, L. salmonis). Controls of L. salmonis (Ls) and C. elongatus (Ce) with mark ladder (M) on both sides of all the sea lice samples. With only a single exception (27, Ce), all bands appeared to have L. salmonis product size.*

3.3 Identification of previous hosts of *C. elongatus*

The possibility for molecular identification of the previous fish hosts of *C. elongatus* was tested on ten lice collected from grey gurnard, lumpfish, and garfish as a pilot study. The identification method used DNA barcoding with adjustments (annealing temperature) to remove double sequences to identify the previous host in the NCBI library.

A saithe transmission experiment was performed to investigate if saithe DNA could be detected in *C. elongatus* after the lice had been attached to Atlantic salmon for different time intervals. In addition, a starvation experiment was performed to study the period of which it is possible to find saithe DNA in the gut content of the lice using the same time intervals as for the saithe transmission experiment.

3.3.1 Molecular taxonomic method

Sequences returned from the sequencing facility were run in BLASTn to identify the previous fish host by comparing the sequence with the NCBI database. BLAST results give a query cover and a percent identity score (0-100%). The query cover is the DNA sequence compared to sequences in the database and informs how much of the query sequence is covered by the target sequence. If the sequence in the database spans over the entire query sequence, the query cover gives a 100% hit. Percent identity tells us how similar the query sequence is to the target sequence based on how many identical nucleotides are in each sequence. High percent identity gives a more significant match (BIOSEQ Bioinformatics Activity, 2021). Two out of ten sea lice samples were recognized as lumpfish in the NCBI sequence library with a query cover of 100.0% and a percent identity between 99.6-100.0%. One sample matched *Caligus belones* with a query cover of 95.0% and a percent identity of 99.8%. The remaining seven sequences resulted in no hits and were unidentified in the database (Appendix F - Table 1F). The hits of *C. elongatus* and *C. belones* in BLAST indicated that the primers used amplified the COI gene of *Caligus* species, which we were concerned about due to the alignment test of the COI sequences of *C. elongatus* and the different fish hosts (Appendix G – Figure 5G). The alignment test showed that the COI sequences between the parasite and hosts were different, but they had a lot of similar areas, and this risked amplifying the parasite COI sequence. Sequences with no result in BLASTn were manually trimmed to remove misleading data from the sequence fragments without further hits (QIAGEN Digital Insights, 2014). A PCR gradient was performed to remove the double sequences most likely caused by concurrent amplification of *C. elongatus* and the fish hosts by increasing the annealing temperature.

3.3.2 PCR gradient to reduce *C. elongatus* amplification

The DNA barcoding results could not find all of the previous hosts of the *C. elongatus* samples investigated. An annealing PCR gradient was made to reduce the amplification of the sea lice to might remove double sequences. The original annealing temperature was 52°C, and the first PCR gradient ranged from 50-60°C. The agarose gel showed strong bands at all temperatures, and the sequence indicated that there still were problems with unwanted amplification of the *C. elongatus* DNA. The primers for the COI sequences bind inadequately to *C. elongatus*. The agarose gel showed no clear pattern of bands in the samples (Figure 16). There were bands at all temperatures except from sample 2 at 60°C, but some were weaker than others. A larger increase in temperature with a new PCR gradient might prevent the primers from binding to the *C. elongatus* DNA and increase the specificity.

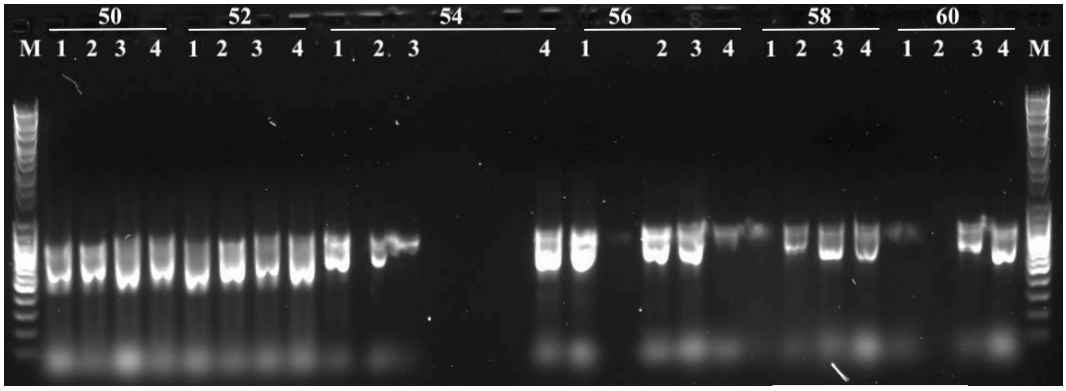

Figure 16. *Agarose gel electrophoresis result of PCR gradient ranging from 50-60°C of four samples C. elongatus. (1 (sample 3), 2 (sample 4), 3 (sample 8), 4 (sample 9)) and mark primers (M) on each side of the samples.*

As the first PCR gradient amplified DNA at all temperatures from 50-60°C, a second attempt was performed with further increased temperatures (58-68°C). Results from the second PCR gradient showed a clearer trend with clear bands at the lowest temperatures and weaker bands at higher temperatures (Figure 17). No bands were detected at 68°C (1-4), and there was no band at samples 4 (64°C) and 2 (68°C).

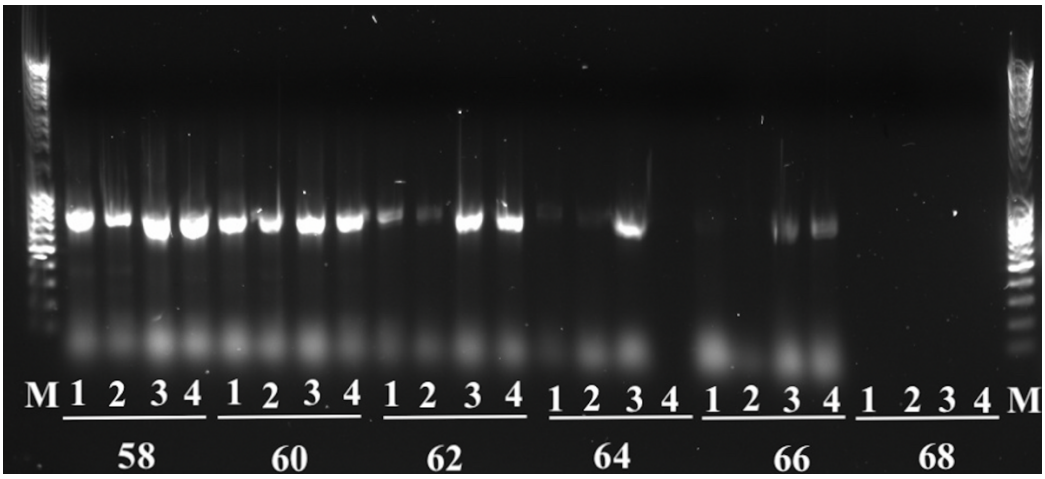

Figure 17. *Agarose gel electrophoresis result of PCR gradient ranging from 58-68°C and four samples of C. elongatus (1 (sample 3), 2 (sample 4), 3 (sample 8), 4 (sample 9)) with mark primers (M) on each side of the samples.*

The samples used in the PCR gradient (3, 4, 8, 9) were sequenced at the sequencing facility with three different annealing temperatures at 52°C, 60°C, and 62°C. Annealing temperature at 52°C was further investigated because it was the highest temperature in the first PCR gradient (50-60°C) which had strong bands on all samples (1-4) (Figure 16). Annealing temperature at 60°C was chosen for the same reason, but for the second PCR gradient (58-68°C) (Figure 17). Annealing temperature at 62°C was chosen as it was the temperature where the samples had detected bands on all samples, but there were weaker bands on samples 1-2 compared to 3-4. The sequencing results received from the sequencing lab were run in BLASTn. Samples 3 and 9 could not find any significant similarities, sample 4 identified *C. lumpus* (the control), and sample 8 at 60°C recognized a bacterium (Appendix F – Table 2F).

The resulting sequences were aligned with the COI gene of their respective previous host (Appendix D) (QIAGEN Digital Insights, 2014). A summary was made of the section of the most similar area of the alignment of the resulting trimmed sequence and the fish's COI gene, and shows the number of bp that matched, mismatched, and was unknown (Table 9). Samples 4 and 9 have a high proportion of 98.6% and 93.7% matched nucleotides, respectively. Sample 8 has the highest percentage of mismatched nucleotides at 42.5%, while sample 3 has 72.2% matches with the highest number of unknown nucleotides (N) at 17.0%.

Table 9. *A summary of the best sections of the alignment of C. elongatus sequence and the host's fish COI sequence from four samples (3, 4, 8, and 9). Information about the number of nucleotides in the section (n) and percentage (%) of bp used in the selected section. The number of matched (Match) and mismatched (Mismatch) nucleotides in the COI sequence, and the number of unknown and not recognized nucleotides (Unknown, N). Sample 4 is highlighted in green as this was the sample where the previous host was recognized in BLASTn (the control).*

Alignment of *C. elongatus* sequence and the hosts fish COI sequence								
Parameter	**Sample 3**		**Sample 4**		**Sample 8**		**Sample 9**	
	n	%	n	%	n	%	n	%
Number of nucleotides	270	98.5	360	46.2	360	50.6	270	41.6
Matched	195	72.2	355	98.6	203	56.4	253	93.7
Mismatched	29	10.7	1	0.3	153	42.5	8	3.0
Unknown, N	46	17.0	4	1.1	157	1.1	9	3.3

3.3.3 Saithe experiments

3.3.3.1 Transmission of C. elongatus from saithe to salmon

Each of the six salmons supplied by the IMR was attempted infected with three sea lice's each. The lice that settled on the salmon were attached to the fish after approximately 15 minutes. Sixteen out of 18 lice from the saithe attached to the surface of the salmon, while two died or did not manage to attach the host. Four lice had detached the fish in the period leading up to sampling. The result from the agarose gel picture shows detection of saithe DNA from the gut contents of *C. elongatus* from the different time intervals attached to a new host (Atlantic salmon) (Figure 18). The first four samples (t1-t4) were attached to the salmon for 1 hour and had strong bands of saithe on the agarose gel. The following four samples (t5-t8) that sat on the salmon for 3 hours had weaker bands of saithe except for one strong band (t7). The remaining four lice (t9-t12) sat on the salmon for 22 hours and had weak bands, except for one sample (t12). The agarose gel was unclear and blurry, but some bands on the agarose gel detected saithe DNA from the *C. elongatus* investigated.

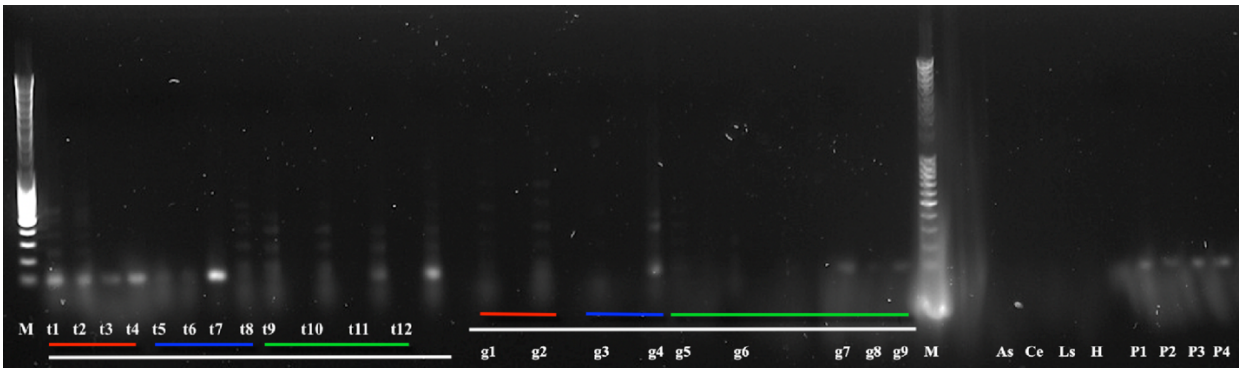

Figure 18. *Agarose gel electrophoresis result of the saithe experiments (transmission- and starvation study) performed on C. elongatus caught in Austevoll, Vestland. Samples with time intervals of 1 hour are highlighted in red, three hours in blue, and 22 hours in green. Sample t1-t12 were transferred from saithe to farmed salmon, and sample g1-g9 was removed from the saithe and starved for different time intervals. A negative control sample of Atlantic salmon (As), C. elongatus (Ce), L. salmonis (Ls), H_2O (H), and four DNA samples of saithe was used as a positive control (P1-P4) (provided by the IMR), and an additional mark ladder (M) on each side of the samples.*

3.3.3.2 Starvation of C. elongatus from saithe

Ten *C. elongatus* was found on a total of six fished saithe hosts, the lice were gently removed from the saithe and put into corning tubes to starve in water for 1, 3, and 22 hours. One louse died before the time interval of 3 hours and was therefore not used in further data. Investigation of the saithe DNA in the gut content of nine *C.* elongatus samples was examined over different time intervals (g1-g9) (Figure 18). The first two samples (g1-g2) removed from the fish to starve for 1 hour had weak bands. The following two samples (g3-g4) were starved for 3 hours. Sample 4 (g4) had a slightly stronger band, and sample 3 (g3) had a weaker band than lice starved for 1 hour. The last five samples (g5-g9) were starved for 22 hours. There was no band from samples 5 and 6 (g5-g6), two bands that were slightly clearer (g7-g9), and there was one weak band (g8).

4 Discussion

4.1 Investigation of *C. elongatus* fecundity

Lice from wild vs. farmed fish

There are no previous studies on whether the origin of the host of *C. elongatus* affects their fecundity. It is important to highlight that all lice from farmed origin were collected from one site at one sampling. The results show that ovigerous lice from farmed fish are significantly smaller than those from wild fish. In the current study, lice from farmed fish were approximately 7% smaller than lice from wild fish. These findings are in line with other studies performed on *L. salmonis*. Tully and Whelan (1993) found significant differences in body size and fecundity between *L. salmonis* lice of farmed and wild origin, where lice on wild salmon were larger and carried twice the number of eggs than lice from farmed fish. In a study from Ireland, Jackson and Minchin (1992) observed significant differences in size between gravid *L. salmonis* from farmed and wild salmon. Lice from wild fish had a higher output than lice from farmed salmon. A study conducted by Nordhagen et al. (2000) found that *L. salmonis* with wild origin were significantly longer and wider than lice from farmed salmon.

There was a significant correlation between the number of eggs and the egg string length for lice from farmed and wild origin. However, the coefficient of determination was lower for lice from wild origin than anticipated, r^2=0.17, meaning that 83% of the variation in the number of eggs for *C. elongatus* from wild origin is explained by other factors than the egg string length. This is dramatically lower than for lice from farmed origin where r^2=0.53. It proved to be a regional difference in the coefficient of correlation for lice from wild fish in south, r^2 = 0.22, and north, r^2=0.14. There was no correlation between lice body length and the number of eggs from lice with farmed and wild origin, which implies that lice size does not influence how many eggs the lice produce.

Lice from wild fish have longer egg strings than lice from farmed origin, and they had approximately the same number of eggs per mm egg string. There was a minor difference of 0.7 eggs per mm egg string which indicates that the size of the eggs is independent of the origin of the lice. The egg string length of lice from wild origin was expected to be longer than for lice from farmed origin as several studies showed that *L. salmonis* from wild origin was longer than lice from farmed salmon (Jackson & Minchin, 1992; Tully & Whelan, 1993). These results correspond well with the results of the current study. The left and right egg strings had the same length and number of eggs for lice with wild and farmed origin.

The number of eggs in the egg strings from farmed and wild hosts of *C. elongatus* was investigated. The box plot for farmed origin has an IQR of 28 eggs compared to wild origin with 50 eggs. Lice from farmed origin had less spread in the data (lower IQR) than lice from wild origin (higher IQR). Lice originating from farmed fish might be exposed to chemotherapeutics impacting the lice´s size and fecundity (Tully & Whelan, 1993).

The average number of eggs is higher for lice from wild origin (130) than lice from farmed origin (95). Tully and Whelan (1993) also found that *L. salmonis* found on wild Atlantic salmon had twice as many eggs as lice from farmed fish. Jackson and Minchin (1992) stated that the origin of the Atlantic salmon impacts the number of eggs significantly, where lice from untreated farmed fish had a lower output than wild fish. Their study showed that *C. elongatus* reproductive output was 54 (Ireland) and 89 (Canada) eggs per egg string with untreated farmed Atlantic salmon as host. The first result is consistent with the results from this thesis. Hogans and Trudau (1989) found that *C. elongatus* carries 89 eggs in each egg string on cultured Atlantic salmon, findings from Ireland correspond well with the results in this thesis, this might be due to that Ireland is closer to Norway than Canada. Pike et al., (1993) found that *C. elongatus* had approximately 80 eggs in the egg strings of cultured rainbow trout (*Oncorhynchus mykiss*). There are few previous fecundity studies on *C. elongatus* with wild origin, this has made it difficult to compare the results with other studies. There is limited research on the topic for other *Caligus* species as well.

Body length and the number of eggs of lice from different hosts

The length of the lice and the number of eggs were examined to investigate if the host species impact the sea lice's growth and fecundity. Less preferred host species, their state of health, and genetic differences may affect egg production (Mackinnon, 1998). There was a significant difference in the number of eggs and the sea lice host, where the number of eggs for *C. elongatus* from Atlantic salmon differed from the other host species. This can be explained by the farmed origin of the Atlantic salmon. To get a better understanding of whether the number of eggs for *C. elongatus* from Atlantic salmon is different from other host species, there should have been collected lice from wild Atlantic salmon. There was a significant difference in the number of eggs between garfish and Atlantic char, and between garfish and lumpfish. The sea lice host with the highest average number of eggs was Arctic char (161), followed by lumpfish (147), trout (144), garfish (123), and farmed Atlantic salmon (95). Lice from Atlantic salmon and garfish have the lowest IQR of the species studied, indicating they have the lowest variance of the number of eggs. There was no significant difference in lice size for different host species.

The length of lice from the different hosts varied by 9% (0.5 mm), but if the lice from Atlantic salmon, which accounted for most of the data from farmed origin are removed, the difference would decrease to 6% (0.36 mm).

The number of eggs and geographical regions

The results revealed a significant difference in the number of eggs from wild *C. elongatus* in different regions in Norway, where lice from southern Norway were different from the other regions. The number of eggs from lice with wild origin in the south was significantly different from central and northern Norway, this may indicate that there is a north-south gradient for *C. elongatus* genotypes 1 and 2. No reports of the number of eggs and the geographical location of wild *C. elongatus* in the Norwegian coast were found, which makes it difficult to compare results with other experiments. However, a fecundity study of *C. rogercresseyi* from farmed fish showed a significant difference in egg string length between different localities in Chile (Bravo, Erranz, & Lagos, 2009). If the study were to be repeated, the genotypes should be confirmed with a larger sample size from sites all along the Norwegian coast

An apparent limitation of the fecundity investigation is the unevenly distributed sampling size, where almost one-third of the data of *C. elongatus* were from farmed Atlantic salmon. Some host species of *C. elongatus* have too few observations that constitute too high uncertainty for the result to be significant. A larger sample size should validate the results.

4.1.1 Duration of *C. elongatus* nauplius stages

The experimental results show that the duration of *C. elongatus* nauplii stages with a temperature of 9.5 ± 0.2°C was 4.03 DPH. The nauplius I stage lasted for 1.64 DPH, and the nauplius II stage lasted for 2.39 days, on average. There was, as expected a significant difference in the duration of the nauplius stages where the duration of nauplius I was longer than nauplius II.

Pike et al. (1993) found that the nauplii stages of *C. elongatus* lasted for 3.99 DPH at a temperature of 10°C. The duration of the nauplius I and II stages were 1.15 and 2.84 days. Another experiment performed by Piasecki and McKinnon (1995) found that the nauplii stages lasted for 3.84 days at a temperature of 10°C, whereas the nauplius I and II stages lasted for 1. 00 and 2.84 days. The duration of the nauplius I stage lasted slightly longer in our results compared to Pike et al. (1993), and Piasecki and McKinnon (1995). It was expected that the duration of the nauplius I stage would be slightly longer in our result as our lice were exposed to a lower temperature, which leads to slower development of sea lice. On the other hand, the

duration of the nauplius II stage was slightly shorter than their consistent results which was not expected. The duration of the naupliar stages was very similar, but our result was somewhat longer as anticipated. Hogans and Trudeau's (1989) found that the nauplius II stage lasted for 1.46 days with the same temperature used in our study, which is in line with our result.

Myhre (2021) examined the duration of the nauplius stages of *C. elongatus* at 9.5°C at the same period as this thesis was written. The results show that the nauplius I stage lasted 1.9 days which was slightly longer than the current results of 1.64 days. The duration of the nauplius II stage lasted 4.8 days, more than twice the current finding of 2.39 days. It was not expected such a large variation in the duration for the nauplius II stage compared to Myhre's (2021) experiment as the lice were exposed to the same temperatures. However, by comparing similar results from other studies, it can be concluded that the experiment was successful as the current study confirmed previous findings.

Small individual and internal differences will always occur, and the deviation from the other experiment might occur due to the lice only being examined once a day. The recordings were not taken at a specific time of the day but were examined between 12:00 to 16:00. However, this was a major score of limitation, and it would be a better solution in terms of finding the accurate duration of the nauplius stages to observe them more frequently. Another limitation is that the incubators did not have a lid, which led to four adult sea lice managing to escape from the incubator. If the study were to be repeated it is recommended with a more frequent observation of the animals at fixed times to get more exact molting times, which gives a more precise duration of the nauplius stages of *C. elongatus*.

4.1.2 Attachment sites of chalimus larvae

Section 3.2.2 investigated the correct sea lice species of the lice that settled on the salmon for 67 out of 136 samples. The results revealed that the infection of *C. elongatus* on the salmon was not successful as only one *C. elongatus* was identified molecularly. The sea lice species molecularly identified accounted for half of the lice examined in the attachment pattern experiment, and it was therefore assumed that the majority of the remaining lice were *L. salmonis*. The infection rate of *C. elongatus* was too low to be able to investigate preferred attachment sites of *C. elongatus*. The attachment pattern is most likely a reflection of where *L. salmonis* chalimi larvae prefer to attach to the salmon.

Several studies have been performed on where sea lice attach to the fish and whether the lice favor specific regions based on the different developmental stages in the life cycle. Bui, Oppedal, Nola, and Barrett (2020) found that *L. salmonis* chalimus II larvae had the highest

abundance on the ventral side of the body (68%), followed by the dorsal side and the head. That is at variance with the present study, where 66% of the lice from the first sampling day were attached to the dorsal side of the fish and 34% to the ventral. The results from the second sampling were also in contrast to their study, where 83% of the lice were attached to the dorsal side and 17% to the ventral. The current study and Bui et al. (2020) had the least number of lice located on the head, based on the zones the fish was divided into. Treasurer and Wadsworth (2004) found the dorsal fin as the primary attachment site with 50% of the chalimus lice of *L. salmonis*. This compares well with the results from both test groups in this study, where the main attachment site was the dorsal fin with 39% and 54% of the lice. Wootten et al. (1982) reported that chalimi *L. salmonis* larvae were commonly found on the dorsal and pelvic fins or around the anus. The results from this thesis provide additional support for similar attachment patterns.

The only identified *C. elongatus* louse in the experiment was found on the posterior abdomen of the fish. However, it is not possible to investigate the attachment pattern of *C. elongatus* explicity because of the lack of data. The attachment pattern of *C. elongatus* was studied by Treasurer and Wadsworth (2004) of farmed Atlantic salmon in Scotland. They found that chalimus larvae of *C. elongatus* were mainly attached to the fins and preferred the pectoral and caudal fin of the fish.

If this experiment were to be repeated, the fish should have been infected with the sea lice species separately in order to increase the probabilities of successfully infecting *C. elongatus*. The co-infection of the lice species was performed because the macroscopical species identification experiment was performed simultaneously with the same materials used in the preferred attachment site experiment. However, it is a known fact that infection attempt of *C. elongatus* in laboratory conditions is challenging to execute, and there are no protocols of how to infect them successfully (S. Dalvin, personal communication). The results from such analyses should be treated with caution, given that our findings are based on a limited number of samples.

4.1.3 Macroscopical identification of chalimus larvae

Atlantic salmon was challenged with similar amounts of *C. elongatus* and *L. salmonis* copepodites. Only one louse (1.5%) out of 67 chalimi larvae molecularly identified were confirmed to be *C. elongatus*. Most of the lice (54) were salmon lice, however, 12 (18%) of the lice were not detectable on the agarose gel, and some of them might be *C. elongatus*.

It was not easy to distinguish *C. elongatus* from *L. salmonis* in the early life stages and the majority of the identifications were made based on whether the lice had a golden-brown or brownish pigmentation on their body. Separating the species became even more problematic than expected because many of the characteristics selected beforehand were not macroscopically prominent enough to distinguish the species. The result showed that 17.9% (12) of the assumptions were incorrect, and the methods used in this thesis to distinguish *L. salmonis* from *C. elongatus* were not optimal. It is possible that some of the lice not detected on the agarose gel were *C. elongatus*. An explanation for the wrong assumptions of the sea lice species might be because the assumptions mainly were based on two characteristics, body color, and eye pigmentation. These features were not prominent enough macroscopically to correctly distinguish the species, as there was a too high proportion of lice incorrectly identified.

There is no previous research on macroscopic differences between the sea lice species, and few researchers have addressed the issue. Previous work has only focused on microscopic differences between the species. However, the results imply that the infection attempt of *C. elongatus* was unsuccessful because the infection rate of the lice on the salmon was very low. Therefore, it is difficult to explain the extent to which it is possible to separate the sea lice species macroscopically, as only one louse was molecularly identified as *C. elongaus*. It is recommended to repeat the experiment to obtain a higher infection rate of *C. elongatus*, a higher number of correct identifications of *C. elongatus* would confirm whether it is possible to separate the species macroscopically. However, the results suggest that the characteristics used to determine the species were not definable enough to correctly separate them macroscopically. It is suggested to use a template of *C. elongatus* and *L. salmonis* to make it easier to detect the differences between the species during the determination if this experiment were to be repeated. Further work must be carried out to establish whether the sea lice species can be determined based on macroscopical differences.

4.2 Identification of previous hosts of *C. elongatus*

4.2.1 Molecular taxonomic method and annealing PCR gradients

Two out of ten fish hosts were identified as the correct host species, one sample was identified as a *Caligus species*, and the remaining five samples were unidentified. The sequences received from the sequencing facility had a lot of double sequences, meaning that there is DNA from several species in the sequencing result. The sequences were trimmed with no further identification results in BLASTn. The COI primer set was used to detected fishes found in the North Sea, but the fish's COI gene was more similar to *Caligus* COI gene than anticipated, meaning that the primers for the DNA barcoding method captured the COI gene of the *Caligus* species in BLASTn. PCR gradients of the annealing temperature were performed to possibly remove unwanted amplification of *C. elongatus* DNA. Sequences received from the sequencing facility with increased annealing temperatures proved to have no effect as the unwanted amplification still was present, this was confirmed by sequences received from the sequencing facility. However, the method has proved it is possible to find DNA from the previous host in the stomach contents of *C. elongaus*.

A limitation of the method is the decomposition time in the intestines of the *C. elongatus*. If it is to be investigated whether it is possible to identify the previous host, e.g., from lice from a fish farm, the sea lice might have been attached to the farmed host for long enough for the lice to decompose the previous host's fish DNA. However, results from the saithe experiment found saithe DNA from the gut content of *C. elongatus* after 22 hours (3.3.3).

4.2.2 Saithe experiments

Saithe is the most abundant wild fish species observed at Norwegian salmon farms (Uglem, Dempster, Bjørn, Sanchez-Jerez, & Økland, 2009), and frequent *C. elongatus* jumping can be expected in such areas. The high abundance of saithe can carry *C. elongatus* that can transfer to the farmed fish (Hemmingsen et al., 2020; Uglem et al., 2009). In addition, a study found that saithe infected with *C. elongatus* transferred from the saithe to Atlantic salmon in laboratory conditions, which suggests a preference of Atlantic salmon over saithe (Bruno & Stone, 1990).

It proved to be challenging to find *C. elongatus* with saithe as host, as it is necessary to fish the right species (saithe) infected with *C. elongatus* at a certain time when the experiment was executed. This led to small sampling sizes, and it would be desirable with a larger sampling size of *C. elongatus* from wild saithe to decrease the margin of errors.

Transmission of *C. elongatus* from saithe to salmon

Lice were transferred from saithe to Atlantic salmon and were attached to the salmon for 1, 3, and 22 hours. The results showed a slight tendency of weaker bands of DNA from saithe on samples that stayed longer on the salmon. There were two exceptions to the trend where samples 4 (t4) and 12 (t12) that stayed on the salmon for 3 and 22 hours, respectively, had a clear band on the agarose gel which shows the presence of saithe DNA in the samples. It was expected that there would be a trend that showed slightly weaker bands of saithe DNA for lice that stayed the longest on the new host as the enzymes in the stomach as the lice decompose the saithe DNA over time. However, there are several factors to consider. It is very doubtful that the lice ate the same amount of food at the same time. There is a high probability that some of the lice had eaten more than others before they were transferred to a new host, which means that lice which just ate were not as hungry as lice with an empty stomach. In addition, there is limited research on the eating pattern of *C. elongatus*. The lice might eat until it is full and then wait until it is starving before the lice eats again, or eat continuously. Sample t12 had a strong band of saithe DNA after sitting on Atlantic salmon for 22 hours. This may be because the lice had eaten a lot of saithe before it was transferred to the salmon, and therefore not grazed on the salmon. However, it was expected a weaker band of saithe DNA after such a long time on the salmon. The agarose gel had quite blurry bands, but it was possible to detect saithe DNA on the gel. There may be several alternative faults that caused the blurry bands on agarose gel, but it might be due to poor agarose loading.

Starvation of *C. elongatus* from Saithe.

Sea lice from saithe were removed from the host to starve for 1, 3, and 22 hours to investigate if it is possible to detect saithe DNA from *C.* elongatus after different time intervals. The results showed that it is possible to detect saithe DNA in *C. elongatus* which starved for all three time intervals investigated. The clearest bands on the agarose gel were from lice starved for 3 hours, followed by two lice starved for 22 hours. The bands from lice starved for 3 hours was clearer than lice starved for 22 hours and 1 hour. This may indicate that lice starving for 22 hours have digested more of the saithe DNA than lice starved for 3 hours. There are weaker bands on the starvation experiment than the saithe's transmission experiment. This may indicate that lice with no access to a host might be stressed as the lice depend on a host for survival. If the lice spend energy looking for a host it might lead to higher energy consumption, which causes the lice to decompose the food faster than lice that have a host available. Another suggestion for a possible cause of weak bands of saithe DNA from samples of lice that had starved for the

shortest time intervals (1, 3 hours) may be that the sea lice had not been eaten from the saithe recently. If this experiment were to be repeated, the sampling size should be larger. In addition, the shortest time intervals should be more scattered (1, 3) to see larger differences in the agarose gel possibly.

5 Conclusion

The number of eggs from female *L. salmonis* did not correlate with the body size. However, there was a positive correlation between the egg string length and the number of eggs. Lice from wild fish were larger and had more eggs than farmed fish. The left and right egg strings had the same length and number of eggs. There was a difference in the number of eggs among the hosts the *C. elongatus* was collected from, where lice from Atlantic salmon had the lowest average number of eggs. This can be explained by the farmed origin of the Atlantic salmon, as lice from farmed fish produce fewer eggs. However, there is no data from *C. elongatus* collected from wild salmon to compare with, which could have given a different outcome. The results from this study imply that *C. elongatus* host does not affect the lice's size. *Caligus elongatus* from wild fish displayed a significant difference in the number of eggs and the lice's region, where lice from southern Norway differed from lice from central and northern Norway, although some of the sites had limited data. Based on the fecundity research, it can be concluded that origin, region, and egg string length had an impact on the number of eggs of *C. elongatus*. Lice host species might impact the number of eggs, but this requires further study. The size of the lice does not impact the number of eggs. The origin of the sea lice's host affects lice and egg string length, while the lice's host species does not affect the size of the lice.

The results from the experiment of the duration of the nauplii stages were consistent with previous research, which reinforces that *C. elongatus* uses approximately four days to develop from newly hatched nauplius I larvae to copepodites at a temperature of 9.5°C.

The attachment pattern of chalimus *L. salmonis* and *C. elongatus* gives an indication of where *L. salmonis* attach the fish as only one *C. elongatus* louse was identified out of the sea lice molecularly investigated. About half of the sea lice that attached to the salmon was molecularly identified, but it is likely that the majority of the remaining sea lice species attached to the salmon were *L. salmonis*. As the infection rate of *C. elongatus* was too low, it was not possible to determine preferred attachment sites of *C. elongatus*. The lice from the different sampling days were attached to similar areas on the fish´s surface. The dorsal fin was by far the most favored area, followed by the posterior back. The fins were preferred above the body surface of the fish.

Macroscopical identification of chalimus *C. elongatus* and *L. salmonis* with the characteristics used in this study proved to be challenging. The characteristics were not prominent enough to separate the species without using a microscope. The infection of *C. elongatus* was not successful and only one louse was molecularly identified out of the sea lice

investigated, this has affected the results. The body pigmentation was concluded to be the most suitable characteristic when distinguishing the species. The method used in the current study is not sufficient enough to distinguish chalimus *C. elongatus* from *L. salmonis* macroscopically based on the characteristics used in this study.

Identifying previous hosts of *C. elongatus* using the molecular "DNA barcoding" method was tested as a pilot study to investigate if the method could identify DNA from the fish host in the gut contents of the lice (known host). Two out of ten samples were correctly identified. Based on the low success rate it is not recommended to identify previous hosts of *C. elongatus* with the method used in the current study. However, the study has shown promising results of identifying fish remnants in the stomach of sea lice. Saithe DNA in *C. elongatus* gut contents was studied for different time intervals of 1, 3, and 22 hours and it can be concluded that DNA from the host of the lice can be found in the gut contents of the lice after 22 hours.

Future research

Further research on *C. elongatus* fecundity should be aimed at genotype 1 and genotype 2 to investigate potential differences in fecundity between the genotypes. It would also be interesting to investigate if there is a north-south gradient of the distribution of the different genotypes. The study should be examined with a larger sampling size from sites all along the Norwegian coast with approximately the same number of observations for each sampling site. A suggestion for further study is to study the alignment of the COI primers of fish host and *C. elongatus* COI to discover the potential of finding previous hosts of *C. elongatus*. In addition, several experiments with different combinations of the primers used might remove the unwanted amplification of *C. elongatus*. This could optimize the method to recognize the fish COI and not amplify *C. elongatus*. Knowledge of previous host species could provide predictability on when the outbreaks occur by monitoring migration and spawning patterns of the respective fish host species. Finally, we hope the effort put into the experiments can provide inspiration and guidance for future research on similar subjects.

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7 Appendices

7.1 APPENDIX A – Chemicals, instruments, and software

Table 1A. *Chemicals, reagents, and suppliers used in the study.*

Chemicals	Supplier
1% Seakem LE agarose	BioNordica AS, Norway
96% Etanol (EtOH)	Kemetyl Norge AS, Norway
Deoxynucleotide triphosphates (dNTP)	Thermo Fisher Scientific, USA
GelRed 10.000X in water	VWR International, USA
Low Electroendosmosis (LE) agarose	Sigma-Aldrich, USA
Magnesium chloride ($MgCl_2$)	Sigma-Aldrich, USA
MassRuler DNA Ladder Mix marker	Thermo Fisher Scientific, USA
MassRuler DNA Ladder Mix, ready-to-use	Thermo Fisher Scientific, USA
Tricane Methanesulfonate (Tricaine-S MS 222)	Syndel, USA
Tris-acetate EDTA buffer	Sigma-Aldrich, USA

Table 2A. *Collection of forward (F) and reverse (R) primers (P) and 3´to 5´sequences used in the study.*

Primer	P	Sequence 3´to 5´
LsF1939	F	gacatagctt tcccccgctta
LsR1941	R	ggcatttcct cgcctgaata
CeF1940	F	ggcatttcct cgcctgaata
CeR2948	R	ccaatatacc taaacaccga
COI-2-LepF1_t1	F	tgtaaaacga cggccagtat tcaaccaatc ataaagatat tgg
COI-2-LepR1_t1	R	caggaaacag ctatgactaa acttctggat gtccaaaaaa tca
COI-2-VF1_t1	F	tgtaaaacga cggccagttc tcaaccaacc acaaagacat tgg
COI-2-VR1_t1	R	caggaaacag ctatgactag acttctgggt ggccaaagaa tca
COI-2-VF1d_t1	F	tgtaaaacga cggccagttc tcaaccaacc acaargayat ygg
COI-2-VR1d_t1	R	caggaaacag ctatgactag acttctgggt ggccraaraa yca
COI-2-VF1i_t1	F	tgtaaaacga cggccagttc tcaaccaacc aiaaigaiat igg
COI-2-VR1i_t1	R	caggaaacag ctatgactag acttctgggt gicciaaiaa ica
Saithe-F	F	gaatcccaat aattttaata gcct
Saithe-R	R	tcgattgctt agtcatcgag a

***Table 3A**. Kits and suppliers used in the study.*

Kit	Supplier
BigDye® Terminator v3.1 Cycle Sequencing Kit	Applied Biosystems, USA
DNeasy® Blood and Tissue kit	QIAGEN, Germany
ExoSAP-IT™ PCR Product Cleanup	Thermo Fisher Scientific, USA
GoTaq® Flexi DNA Polymerase	Promega Corporation, USA

***Table 4A.** Software and suppliers used in the study.*

Software	Supplier
BLAST®	PubMed Central
ImageJ version 1.8.0	Public Benefit Corporation
RStudio	RStudio Inc.
QIAGEN CLC Genomics Workbench	QIAGEN Inc.

***Table 5A**. Instruments and suppliers used in the study.*

Instrument	Supplier
3730xl DNA Analyzer	Applied Biosystem, UK
Canon EOS 2000D 18-55MM	Canon, Japan
Centrifuge 5415R	Eppendorf, Germany
Electrophoresis Power Supply EPS-300	Pharmacia Biotech, Sweden
GeneAmp PCR system 9700	Applied Biosystems, UK
iBright CL 1000 Invitrogen Imaging Systems	Thermo Fisher Scientific, USA
MS3 vortexer	IKA, Germany
NanoDrop® ND-1000	Thermo Fisher Scientific, USA
Sub-Cell® GT Agarose Gel Electrophoresis Systems	Bio-Rad, USA
Thermomixer ® C	Eppendorf, Germany

7.2 APPENDIX B – Presumption of sea lice species

Table 1B. *Assumed of species of 67 lice based on morphological characteristics (Table 2) of C. elongatus and L. salmonis. The number of lice (Lice nr.) Assumed species (Assumed) and the PCR result of species (PCR). The species examined are L. salmonis (LS) and C. elongatus (CE). L. salmonis that erroneously was thought to be C. elongatus is highlighted with red color, L. salmonis that was identified correctly is highlighted in green. The only C. elongatus of the sample is highlighted with yellow. The lice numbers where the agarose gel bands were weak enough that the lice species were not detectable is marked (-).*

Assumed of lice species								
Lice nr.	**Assumed**	**PCR**	**Lice nr.**	**Assumed**	**PCR**	**Lice nr.**	**Assumed**	**PCR**
1	LS	LS	24	LS	LS	47	LS	LS
2	LS	LS	25	LS	LS	48	LS	LS
3	LS	-	26	CE	LS	49	LS	LS
4	LS	LS	27	CE	CE	50	LS	-
5	LS	LS	28	LS	LS	51	LS	-
6	CE	LS	29	LS	LS	52	LS	LS
7	LS	LS	30	LS	LS	53	LS	-
8	LS	LS	31	LS	LS	54	LS	-
9	LS	LS	32	LS	LS	55	LS	-
10	CE	LS	33	CE	LS	56	LS	LS
11	LS	LS	34	LS	LS	57	LS	-
12	LS	LS	35	CE	LS	58	CE	LS
13	LS	LS	36	LS	LS	59	LS	-
14	CE	LS	37	LS	LS	60	LS	LS
15	LS	LS	38	LS	LS	61	LS	-
16	LS	LS	39	LS	LS	62	CE	LS
17	CE	LS	40	CE	LS	63	LS	LS
18	LS	LS	41	LS	LS	64	LS	LS
19	LS	LS	42	LS	LS	65	LS	LS
20	CE	LS	43	LS	LS	66	LS	-
21	LS	LS	44	CE	LS	67	LS	-
22	LS	LS	45	LS	LS			
23	LS	LS	46	LS	-			

7.3 APPENDIX C – DNA concentration and purity

Spectrophotometer results. The values between 1.8-2.2 ng/ µL with wavelengths of 260/280 nm and between 2.0-2.4 ng/ µL with wavelengths of 260/230 nm are preferred and considered high quality. Abnormal values indicate that the sample is not optimally cleaned. The quality of the DNA purification was high and could be used for further PCR examinations.

Table 1C. *Spectrophotometer results of ten samples with three different host species. DNA concentration (ng/µL), amount DNA (µg) and wavelengths of 260/280 (ng/ µL) and 260/230 (ng/µL).*

DNA extraction				
Sample nr.	**Host species**	**DNA con.**	**A_{260}/A_{280}**	**A_{260}/A_{230}**
1	Grey gurnard	31.02	2.10	2.04
2	Grey gurnard	259.78	2.18	2.39
3	Lumpfish	291.25	2.15	2.38
4	Lumpfish	147.27	2.16	2.50
5	Lumpfish	250.95	2.17	2.37
6	Lumpfish	177.00	2.14	2.12
7	Garfish	215.84	2.22	2.37
8	Garfish	330.71	2.20	2.44
9	Atlantic salmon	278.33	2.15	2.25
10	Atlantic salmon	284.58	2.18	2.38

7.4 APPENDIX D – COI sequences of host species and *C. elongatus*

***Table 1D**. The COI sequence and query of C. elongtus found in NCBI as "Caligus elongatus voucher MT08916 cytochrome oxidase subunit 1 (COI) gene, partial cds; mitochondrial".*

The COI sequence of *C. elongatus*	
Query	**Sequence**
1	aactctttac ttaattagag gattttgatc tgggctggta gggttagcta taagtgttat
61	tattcgttta gaactgtctc aaccaggcct ttatctagga gactcacaag tatataatgt
121	aattgtaact gcccatgctt ttattataat ttttttata gttatacctg tgttaattgg
181	gggatttggt aattggttag tgcccctatt actgggtgcg ccagatatgg catttcctcg
241	cctgaataat ataagttttt gatttttgat gccgtcacta acactactac ttttaagggc
301	tcttgttgaa aggggtgcag gtacagggtg aacagtttac cctcccctat cttctggtgt
361	attccactct ggtgcatcag tagattttgc tattttctct cttcatttgg caggaatttc
421	ttctctttta ggggcggtga attttatcag tacaattctc aatcttcggt gtttaggtat
481	attggttgaa cgaataccta tattcccctg atctgtgctt attaccgccg tattactcct
541	attatcttta cccgttttgg caggagctat tactatacta ttaactgatc gtaatttaaa
601	taccaggttt tttgatccca gtgggggggg ggatcctatt ctctaccaac atttattt

***Table 2D**. The COI sequence and query of lumpfish found in NCBI as "Cyclopterus lumpus voucher HLC-10866 cytochrome oxidase subunit 1 (COI) gene, partial cds; mitochondrial".*

The COI sequence of lumpfish	
Query	**Sequence**
1	cctttatcta gtatttggtg cttgagccgg aatggtcggg acaggcctaa gccttttaat
61	ccgggccgag ctaagccaac ccggggccct cttgggcgac gaccaaattt acaacgttat
121	tgttacggct catgctttcg taataatttt ctttatagta ataccaatca taattggggg
181	ctttggaaat tgactcatcc ccctaataat cggcgccccc gatatagcat tccctcgaat
241	aaacaacatg agtttttgac ttttaccccc ttctttccta ttgcttcttg cctcttcggg
301	cgtcgaagca ggggccggaa ccgggtgaac cgtctaccct cctttagcag gtaacctggc
361	acacgccggg gcctctgtcg acttaacgat cttttcttta cacctcgcgg gaatctcttc
421	aatcctcgga gcaattaatt ttattacaac tatcatcaac atgaaacccc ctgctatgtc
481	ccagtaccag actcccctat ttgtgtgatc tgtccttatt actgccgtac tactacttct
541	ctccctccct gtccttgccg ctggcattac aatgctacta acagaccgca accttaacac
601	caccttcttc gacccagcag ggggcgggga ccccattctt taccaacatc tc

***Table 3D**. The COI sequence and query of garfish found in NCBI as "Belone belone voucher MT02816 cytochrome oxidase subunit 1 (COI) gene, partial cds; mitochondrial".*

The COI sequence of garfish	
Query	**Sequence**
1	cctttatcta gtatttggtg cttgagctgg aatagtgggc actgctttaa gccttcttat
61	tcgagcagaa ctaagccaac caggctctct tctgggtgat gatcaaattt ataatgttat
121	cgtcacggca catgccttcg taataatttt ctttatagta ataccaatta tgattggcgg
181	ttttggaaac tgattaatcc ccctaataat tggagcccct gatatagcat tccctcgaat
241	aaataacata agtttttgat tattaccacc atcattcctc cttcttttag catcatctgg
301	ggttgaagct ggtgccggaa ccggatgaac tgtttacccc cctctagctg gtaacttagc
361	ccacgcggga gcatccgttg atttaacaat tttttctctt catctagcag gtatttcatc
421	aattttaggc gctattaatt ttattaccac tattattaat ataaaaccac ctgcaatttc
481	acaatatcaa accccactat ttgtttgagc cgtattaatt acagccgtcc ttcttctctt
541	atccctaccc gtcctagctg ctggaattac aatacttctg acagaccgaa acctaaacac
601	tacctttttt gatcctgctg gcggtggaga tcctattctt taccaacatt tg

***Table 4E.** The COI sequence and query of grey gurnard found in NCBI as "Eutrigla gurnardus voucher CSFOM-255 cytochrome oxidase subunit 1 (COI) gene, partial cds; mitochondrial".*

The COI sequence of grey gurnard	
Query	**Sequence**
1	tttggtgcct gagctggtat agtaggcaca gccctaagcc ttctcatccg ggcagagcta
61	agccagcccg gtgccctttt aggggacgac caaatctata acgtcattgt tacagctcat
121	gccttcgtaa tgattttctt tatagtaatg ccaatcatga ttggaggctt cggaaactga
181	ctcatcccct tgatgattgg tgcccctgat atggcctttc ctcgaataaa caacataagt
241	ttttgacttc tgcccccttc cttcctactc cttctagcct cctctggggt tgaagccggt
301	gccgggacag gatgaactgt ctaccctccc ttggccggca acttagccca tgccgggggcc
361	tctgtagacc taactatctt ctcccttcat ctggccggga tttcctcaat ccttggtgca
421	attaatttca tcacaaccat tattaatatg aaacctcccg caatctccca ataccagacc
481	cccctgttcg tgtggtccgt gctaattacc gccgtcctcc ttctactgtc cctaccggtc
541	cttgccgcag gcatcacaat gcttcttaca gaccgtaacc taaacaccac attcttcgac
601	cctgccggag ggggagaccc cattctctac caacatcttt tc

7.5 APPENDIX E – Attachment sites of sea lice on Atlantic salmon

Table 1E. *The distribution of number of C. elongatus and L. salmonis on nine Atlantic salmon. The fish host is partitioned into head (H), anterior back (AB), posterior back (PB), posterior abdomen (PA), anterior abdomen (AA), dorsal fin (DO), adipose fin (AD), caudal fin (CA), anal fin (AN), caudal fin (PV) and pectoral fins (PC).*

The distribution of *L. salmonis* and *C. elongatus* on infected Atlantic salmon.

Fish nr.	H	AB	PB	PA	AA	DO	AD	CA	AN	PV	PC	Total nr.
1	-	-	-	-	-	3	-	-	2	-	1	6
2	1	-	4	-	-	3	-	-	1	-	1	10
3	-	-	3	-	-	9	-	-	-	-	-	12
4	-	-	2	1	1	11	1	-	2	1	1	20
5	-	1	3	3	-	7	-	-	-	1	3	18
6	1	1	3	1	2	3	1	-	3	1	-	16
7	-	-	3	-	-	10	1	1	-	2	-	17
8	-	1	3	1	-	4	1	-	-	-	-	10
9	-	1	6	5	-	10	2	1	1	-	1	27
Total nr.	2	4	27	11	3	60	6	2	9	5	7	136

7.6 APPENDIX F – DNA sequences and BLAST results

Table 1F. *The sequence results received from the Seq Lab of the pilot study. Sample number (1-20) (Nr.), forward(F) or reverse (R) primers (P), sequence, blast results with query cover, and percent identity of the samples. Samples with no significant similarity found are removed from the table. Green and red color highlights are positive and negative BLAST results on the previous host of C. elongatus.*

Nr.	P	Sequence	Blast results
4	F	nnnnnnnnnn nnnnnngggn ncnnttnatn tantanttgg nngnttganc ncggaatggt cgggacaggn ctaanccttt taatccgggc cganctnanc caacccgggg ccctcttggg nnacnaccaa atttacaacg ttattgttac ngctcatgct ttnntnataa ttttctttata gtaataccan nnnnaatcgg gggntttgga aattgactca tccccctaat aatcggcgcc cccgatatag cattccctcg aataaacaac atgagtttttg acttttaccc ccttctttcc tattgcttct tgcctcttcg ggcgtcnaan caggggccgg aaccgggtga accgtctacc ctcctttanc aggtaacctg gcacacgccg gggcctctgt cgacttaacn atcttttctt tacacctcgc gggaatctct tcaatcctcg gagcaattaat tttattacaa ctatcatcaa catgaagccc cctgctatgt cccantacca gactcccta tttgtgtgat ctgtccttat tactgccgta ctactacttc tctccctccc tgtccttgcc gctggcatta caatgctact aacagaccgc aaccttaaca ccaccttctt cgacccagca ggggggcgggg accccattct ttaccaacat ctcttttgat tctttggcca cccagaantc	***Cyclopterus lumpus voucher NRM:50348 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial*** Query cover: **82.0%** Percent identity: **95.4%**
6	F	ttattgttac ggctcatgct ttcataataa ttttctttat agtaatacca atcataatcg ggggctttgg aaattgactc atccccctaat aatcggcgcc cccgatatag cattccctcg aataaacaac atgagttttt gacttttacc cccttctttc ctattgcttc ttgcctcttc gggcgtcgaa gcaggggccg gaaccgggtg aaccgtctac cctcctttag	***C. lumpus isolate EJ13 cytochrome c oxidase I (COI) gene, partial cds; mitochondrial*** Query cover: **100.0%** Percent identity: **99.6%**
14	R	gtggtgttaa ggttgcggtc tgttagtagc attgtaatgc cagcggcaag gacagggagg gagagaagta gtagtacggc agtaataagg acagatcaca caaatagggg agtctggtac tgggacatag caggggggctt catgttgatg atagttgtaa taaaattaat tgctccgagg attgaagaga ttcccgcgag gtgtaaagaa aagatcgtta agtcgacaga ggccccggcg tgtgccaggt tacctgctaa aggagggtag acggttcacc cggttccggc ccctgcttcg acgcccgaag aggcaagaag caataggaaa gaagggggta aaagtcaaaa actcatgttg tttattcgag ggaatgctat atcgggggcg ccgattatta gggggatgag tcaatttcca aagcccccga ttatgattgg tattactata aagaaaatta ttacgaaagc atgagccgt	***C. lumpus voucher MT08360 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial*** Query cover: **100.0%** Percent identity: **100.0%**
17	R	aaacaggtaa ggatagtagt agaagaactg ctgtaattaa aacagatcaa ggaaatatag gtatccgctc aactagtatc cctaggcaac gtaaatttaa aatagttcta ataaaattta ctgcacctaa tagagacgaa acgcctgcta agtgtaaaga aaaaatcgca aaatctactg aagcacctga atgaaacacc ccagaagata agggagggta aactgtccac cctgtgcccg ctccccttc tacaagagcc cttagaagta gtagggttaa tgaaggcatt aaaaatcaaa atctcatatt attgagtcga ggaaaagcta tatcaggagc ccctaataaa agagggacta gtcaattccc aaatccccca atcaacacag gcataaccat gaaaaaaatt atgataaaag catgggctgt aacaataaca ttatacactt gggagtctcc caaatatatt ccaggttggg ataattccaa tcgaataata actcttatag ctaatccgac aagcccggat	***Caligus belones cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial*** Query cover: **95%** Percent identity: **99.8%**

Table 2F. *The sequence results after the first temperature gradient. Four samples were examined with an annealing temperature at 52°C (41, 42, 43, 48). The table includes the sample nr., template nr., PCR annealing temperature, previous host of the louse, the original sequence, the cleaned sequence performed in CLC, and BLAST results with additional information about percent identity and query cover. Correct matches are highlighted in green color.*

Sample	Sequence	Trimmed sequence	BLAST results
Template: **3** Temp: **52** Host: **Lumpfish**	nnnnnnnnnn nngggnnctc ttnacntnnn ntaggannnt tgancnggnn tggtnnggnt anctntaann gttnnnattc ggttagaact gacncaaccc ggnnttnann tnnganactn nnaantntac aatgtaattg tnacngccca tgcttttnnn ataatttttt ttatagttat accngtgtta attgggggat ttggnaattg aatnatgnnc ctnntnnnn nnncccncn nnnntngcnn tcccnnnna nnaacnannn nnnnttttga nttttanncn cctcatnnnn nnnnnnnnnn nnnnan	ttgatctggg ntggtngggn tagctntaan ngttnntatt cggttagaac tgacncaacc cggnnttnan ntngganact nncaantnta caatgtaatt gtaacngccc atgcttttan nataattttt tttatagtta taccngtgtt aattggggga tttggnaatt gaatnatgnn cctnntcgtn nnncccnccn anntngcnnt cccncnaann aacnananga ntttttgant tttacncncc tcat	***No significant similarities found.***
Template: **4** Temp: **52** Host: **Lumpfish**	nnnnnnnnnn nnnnnngggn ncnnttnatn tantanttgg nngnttganc ncggaatggt cgggacaggn ctaancctttt taatccgggc cganctnanc caacccgggg ccctcttggg nnacnaccaa atttacaacg ttattgttac ngctcatgct ttnntnataa ttttctttata gtaataccan nnnnaatcgg gggntttgga aattgactca tccccctaat aatcggcgcc cccgatatag cattccctcg aataaacaac atgagtttttg acttttaccc ccttctttcc tattgcttct tgcctcttcg ggcgtcnaan caggggccgg aaccgggtga accgtctacc ctcctttanc aggtaacctg gcacacgccg gggcctctgt cgacttaacn atcttttctt tacacctcgc gggaatctct tcaatcctcg gagcaattaat tttattacaa ctatcatcaa catgaagccc cctgctatgt cccantacca gactcccta tttgtgtgat ctgtccttat tactgccgta ctactacttc tctccctccc tgtccttgcc gctggcatta caatgctact aacagaccgc aaccttaaca ccaccttctt cgacccagca gggggcgggg accccattct ttaccaacat ctcttttgat tctttggcca cccagaantc tagtcatann nnntnnccng nnnnnnnnna cncatctngn gattctttgg ccacccanaa gtctantcat agctgttnnc ngnn		***Cyclopterus lumpus voucher NRM:50348 cytochrome oxidase subunit I (COI) gene, partial cds; mitochondrial*** Query cover: **82.0%** Percent identity: **95.4%**
Template: **8** Temp: **60** Host:	attctatacc tgttcgtagc cggttttgtc ggattcttgt ccgtcatgtt cacagtttac atgcgcatgg agctaatgaa ccctggtgtt caatacatgt gtatggaagg cgcgcgtctg ttccctgctg cgctcgacga atgtacacct aacggccacc tctggaacgt gatgatcacg taccacggcg tgctaatgat gttctttgta gttattcctg cgcttttcgg cggctttggt aactacttca tgccactgca aatcggtgcg cctgacatgg cattcccgcg tttgaacaacc	ctatacctgt tcgtagccgg ttttgtcgga ttcttgtccg tcatgttcac agtttacatg cgcatggagc taatgaaccc tggtgttcaa tacatgtgta tggaaggcgc gcgtctgttc cctgctgcgc tcgacgaatg tacacctaac ggccacctct ggaacgtgat gatcacgtac cacggcgtgc taatgatgtt ctttgtagtt attcctgcgc ttttcggcgg ctttggtaac	***Octadecabacter temperatus strain SB1, complete genome***

Garfish

tgtcattctg gctgttctgt actggtgtcg cgcttggtgt ttcttccctg ctcgcaccag gtggtaacgg tcagctcggt tccggtgttg gttgggttct gtacccgccg ctctccacaa ctganacagg cttttcaatg gacctcgcga tcttcgcagt tcacgtctct ggtgcgtctt ctatccttgg cgcaatcaac atgatcacaa ccttcctgaa catgcgcgcc cctggtatga cgctgcacaa agtgccgctg tttgcttggt cgatcttcgt taccgcangg ctgatccttc tggctctgcc tgttctggct ggcgcaatca caatgctttt gatggaccgt aacttcggta caaccttctt tgaccctgct ggcggcggnn accctgtccc tttaccagca catcttgtga ttctttggcc acccaaaaat ctagtcnta

tacttcatgc cactgcaaat cggtgcgcct gacatggcat tcccgcgttt gaacaacctg tcattctggc tgttctgtac tggtgtcgcg cttggtgttt cttccctgct cgcaccaggt ggtaacggtc agctcggttc cggtgttgg ttgggttct gtacccgccg ctctcca--- ---caactga nacaggcttt tcaatggacc tcgcgatctt cgcagttcac gtctctggtg cgtcttctat ccttggcgca atcaacatga tcacaacctt cctgaacatg cgcgcccctg gtatgacgct gcacaaagtg ccgctgtttg cttggtcgat cttcgttacc gcanggctga tccttctggc tctgcctgtt ctggctggcg caatcacaat gcttttgatg gaccgtaact tcggtacaac cttctttgac cctgctggcg gcggnnaccc tgtcccttt accagcacat

Query cover: **99.0%**

Percent identity: **88.7%**

Template: **9**

Temp: **60**

Host:

Garfish

gnnnnctana cnnnnnnntn gnngnnnnnn ncggattcnn nnnnnnnnng ntnnnnctttt nnatgcnnnn ggaannnang nnncctggng nnnnnnnnnn nnannangaa ngnnngnnnn ngttnnnngn nncnntnnnn nantgnnnnn tnnnnnnnac ntctggaann nnnnnannnc nnncnnnggn nngnnnntna tnnnnnttgt nnnnntgccn gcnnnnntcn nnnncnnnnn nannaannnc ntgnnnntnn naattnnnnc nccngnnntg nnnctccnnn gnnngaacnan nngnnnnnnn nnnnnnncng nacngganna ncnntngann nnnctncnnn nnnnnnncnn ggngnnaacn gnnnnnnnnn nnnnnnnnnn nnngnnnnnc nnnnnnnnnn nnnntnnnnt nnnnnnnngn nnnnnnnnnn nnntnnnnnn nttnnnantt nannncncnn gnnnntctnn nannntnnnn ncnnncnnnn tnnnnnnnnn nnnnnnnann nngcncgccn ntngnntnac nctnnncnnn ntnncnnnnn nngcntgnnn nannnnnnnt accgacnnga nntgannnnn ctgnntntnn ntgnnncnnn nngnnnnann anannnnnct nttnnnatnn acnnnnnnnn nngntncnnc cnncnnnnna nnnnnntngn nnnnnnnnnn nnnnnngnnt taccancaca tcttgtgatt ctttggncacc canaantcta gtcntannnn ntttncngnnn

ggnnngnnan tnatnnnnnt tgtantnntgc cngcnnnntt cgnnnncnnn ngnannaann ncatgncnnt nnnaattnnn ncnccngncn tgnnnctccn ncgttngaac nannngnnnn nntnnnngan cngnacngga nnancnctng annnnnctnc nctgnncnnn cnnggngnna acggacntnn cgntnncnnn gttnnttgnn nncnnnnnnn gcngntntnn cntcnnnnnn ggnnnntnan nnnnnntnnn nnnnttnnna attnannncn cnngnnnntc tnnnannntn nnnncnnncn nnntgatnna ncnnttnnnn annntgcncg cccntngtat nacnctncnc nnngtgccnc tgnnngcntg nncnanctnc tttaccgacn ngaantgan nntnctgnnt ntnnntgann cnggcngnnn nannanannn nnctntttgn atnnaccnnn ncttnngntn cnnccntcnn nnnannnngc tngnnnnagn nnnnnnnnnn gnnttaccan cacatcttgt gattctttgg ccaccca

No significant similarity found.

7.7 APPENDIX G – Alignment of samples and previous host

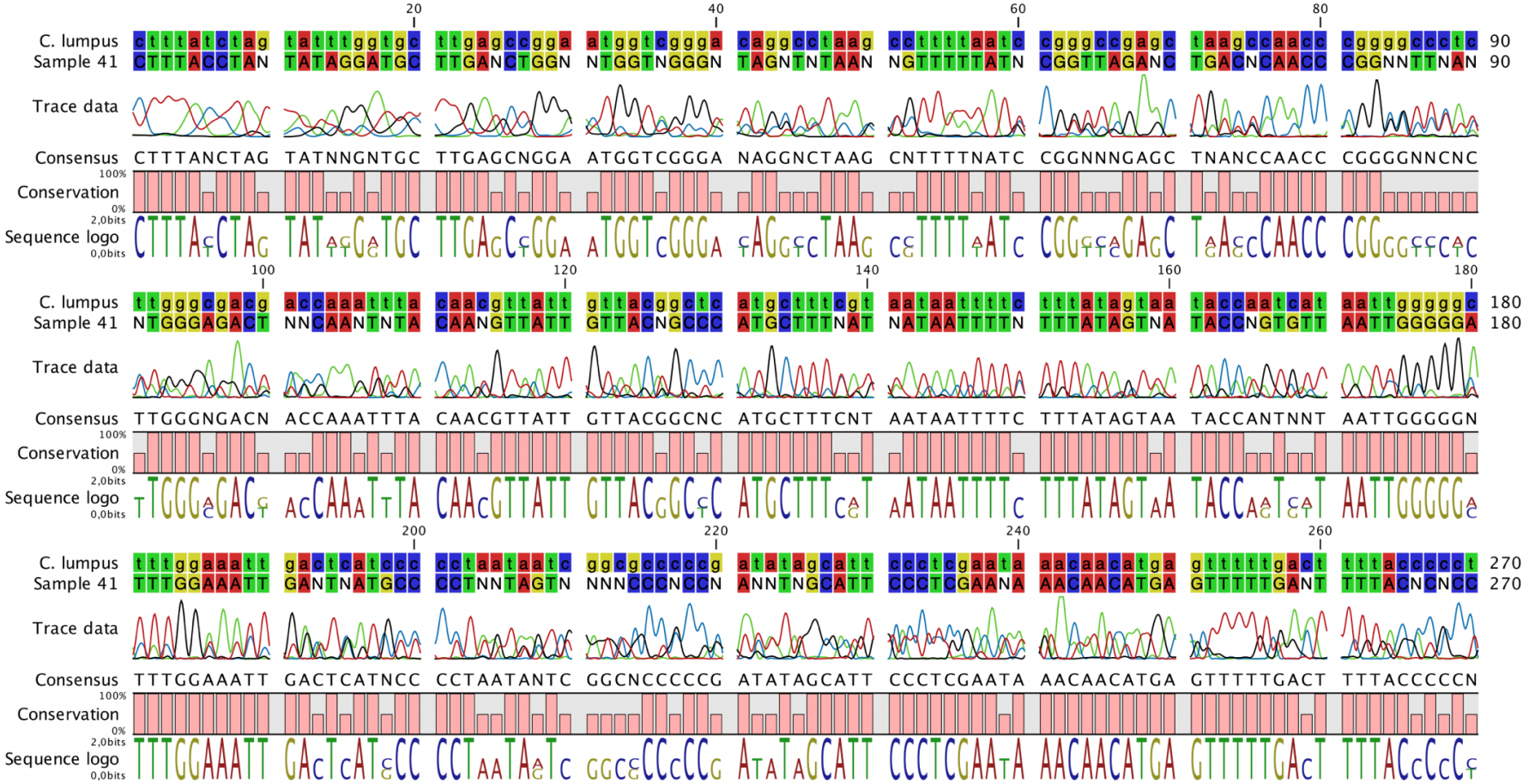

Figure 1G. *A fragment of the alignment of sample 3 and the COI sequence of lumpfish from CLC. Sample 3 had a short sequence, an annealing temperature of 52 °C, and C. lumpus were prior host.*

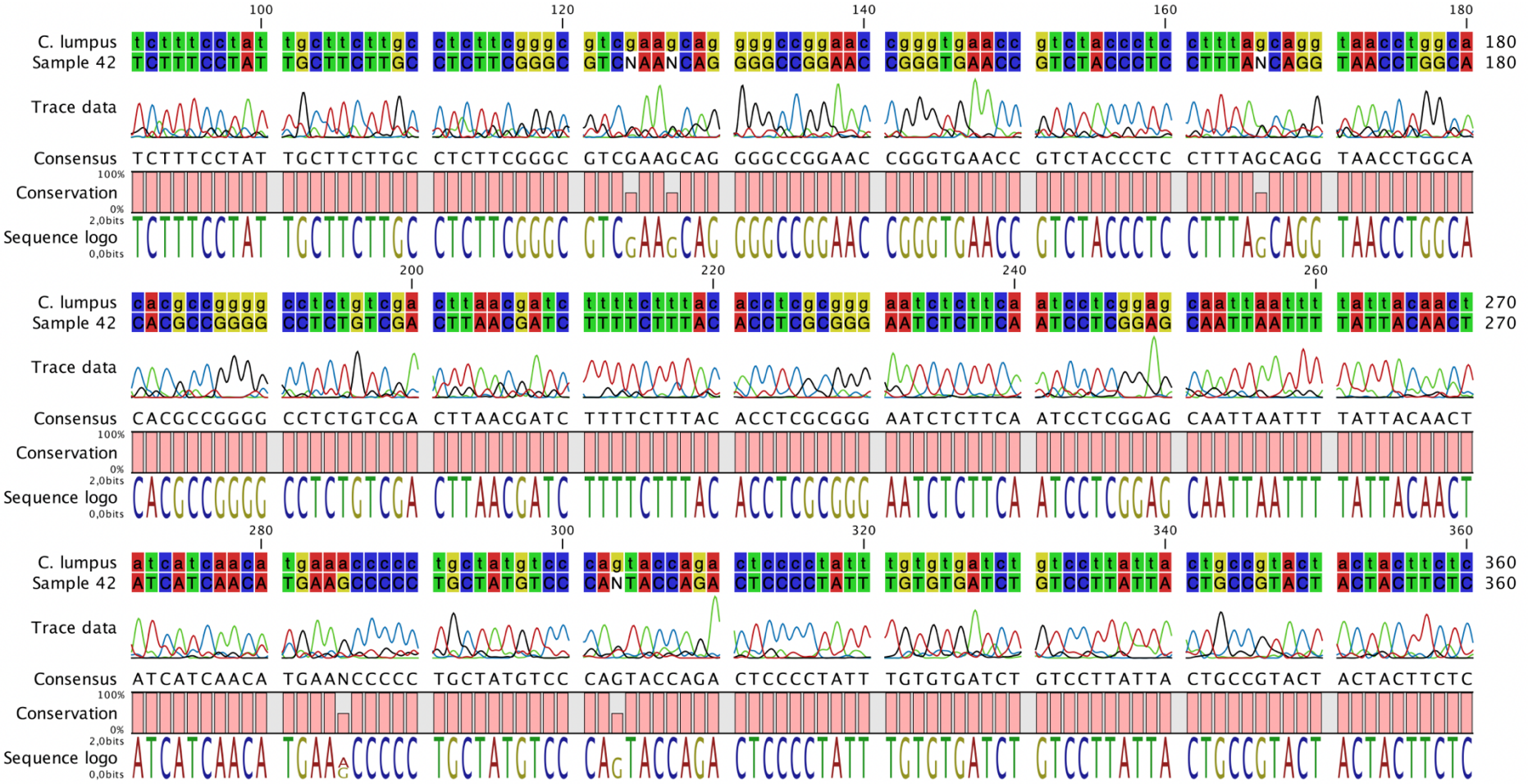

Figure 2G. *A fragment of the alignment of sample 4 and the COI sequence of lumpfish from CLC. Sample 4 had a good sequence, an annealing temperature of 52 °C, and C. lumpus was the prior host.*

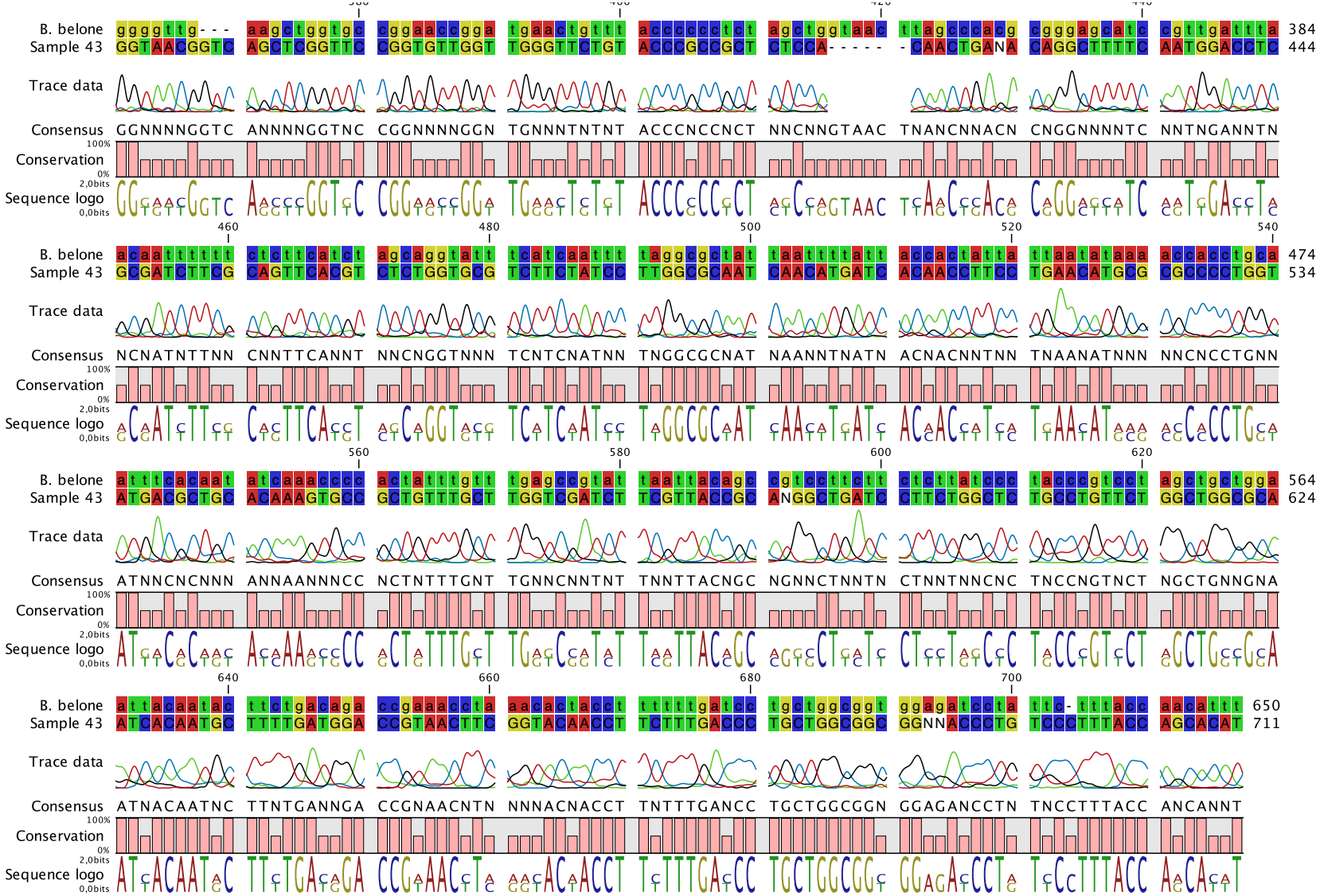

Figure 3G. *A fragment of the alignment of sample 8 and the COI sequence of garfish from CLC. Sample 8 had a bad sequence, an annealing temperature of 52 °C, and B. belone was the prior host.*

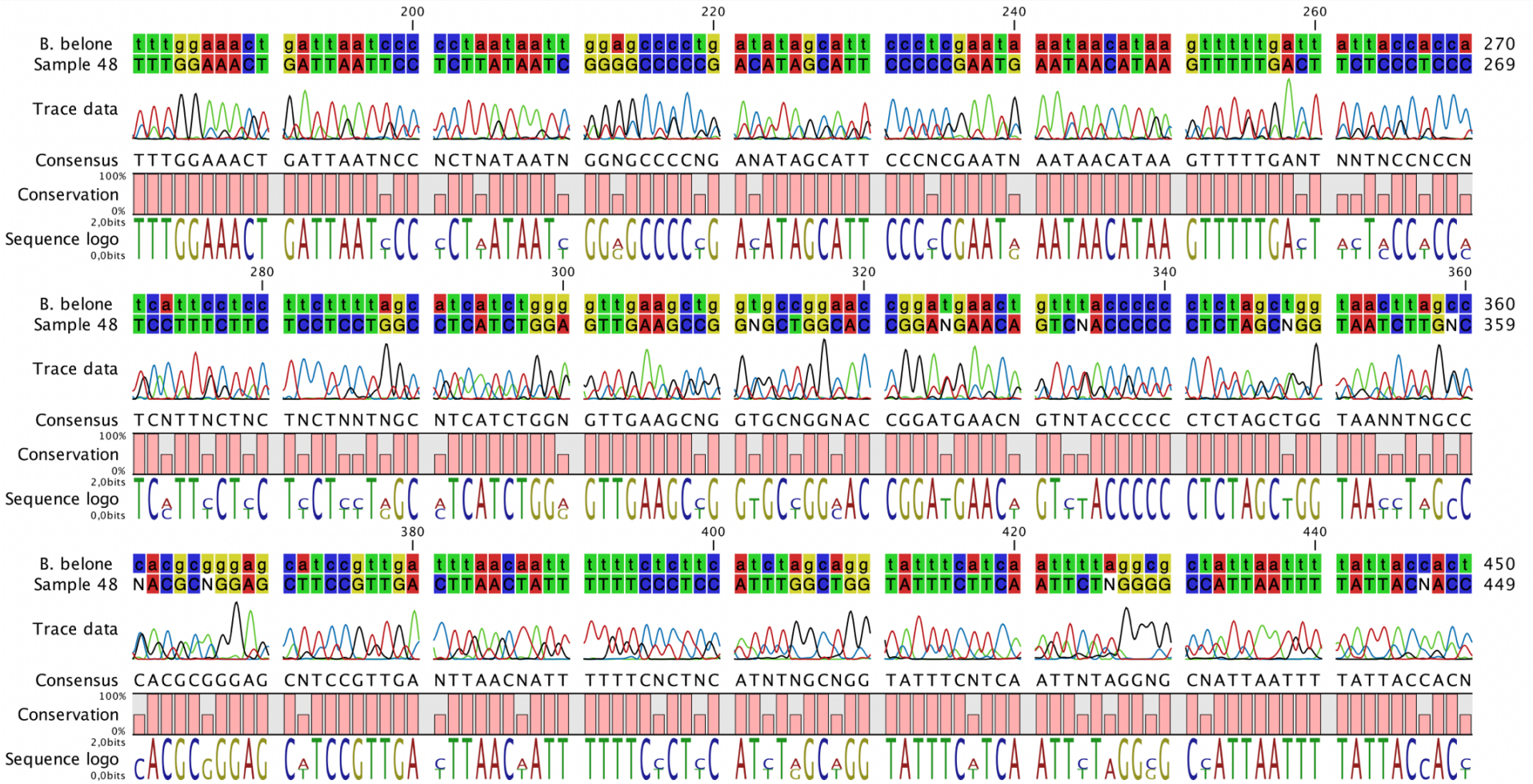

Figure 4G. *A fragment of the alignment of sample 9 and the COI sequence of garfish from CLC. Sample 9 had many double sequences, an annealing temperature of 60 °C.*

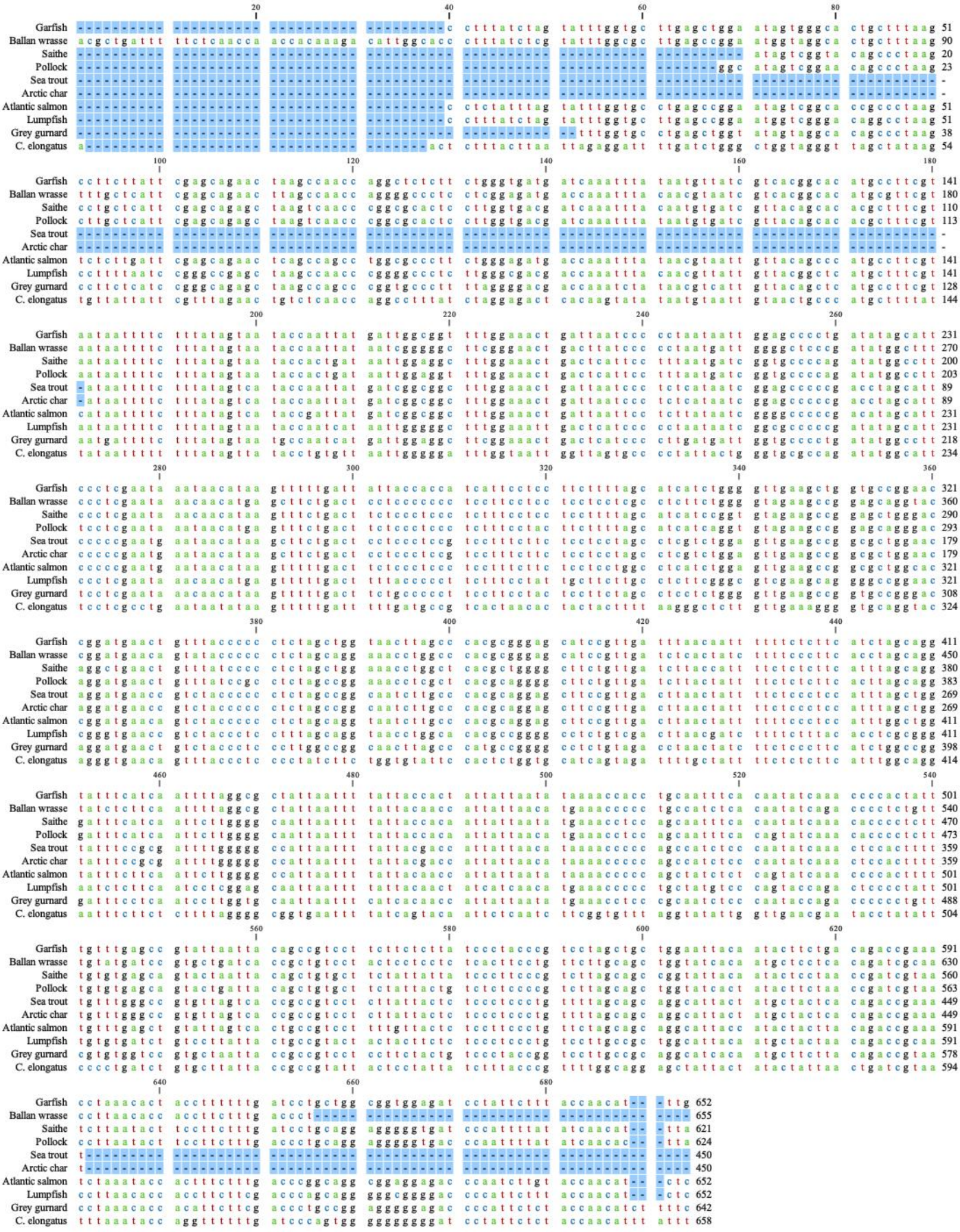

Figure 5G. *Alignment of the COI sequence of lumpfish, grey gurnard, garfish, and C. elongatus from CLC.*

7.8 APPENDIX H – Statistical analysis

RStudio script

```
library(tidyverse)
library(RColorBrewer)
library(extrafont)
library(ggpmisc)
library(readxl)
library(dplyr)
library(gridExtra)

setwd("~/Desktop/R")

data <- read.csv("Fecundity.csv", header = TRUE, sep = ";", dec = ",") %>% as_tibble() %>%
  filter(Host %in% c("Arctic char","Trout","Garfish","Lumpfish", "Atlantic salmon")) %>%
  mutate(origin = case_when(
    Locality== "Låva" ~ "Farmed",
    TRUE ~ "Wild"))


# 1 Linear regression models of the number of eggs compared to the egg string- and sea lice length.

Silje <- c("#a4d3ee", "#36648b")

my.formula <- y ~ x

plot1 <- data %>%  as_tibble() %>%
  select(Lice.L, Av.egg.string.L, Total.eggs, origin) %>%
  rename("b) Body length" = Lice.L, "a) Egg string length"=Av.egg.string.L) %>%
  pivot_longer(cols = c("a) Egg string length", 'b) Body length'),
               names_to = "Average.length", values_to = "Value") %>%
  ggplot(mapping=aes(x=Value, y=Total.eggs, colour=origin)) +
  geom_point() +
  geom_smooth(method = "lm", level=0.95, alpha=0.3) +
  #scale_color_brewer(palette="Paired") +
  scale_colour_manual(values=Silje)+
  theme_bw() +
  theme(text=element_text(size=20,  family="Times New Roman"), strip.text.x = element_text(size = 25)) +
  labs(x="Length (mm)", y="Total number of eggs", colour="Origin") +
  stat_poly_eq(formula = my.formula,
               eq.with.lhs = "italic(hat(y))~`=`~",
               aes(label = paste(..eq.label.., ..rr.label.., sep = "~~~")),
               parse = TRUE) +
  facet_wrap(~Average.length, scales = "free_x")

plot1

ggsave("length_nreggs.png", plot1, height = 7, width = 11)


# Calculations of the average and the median number of eggs and egg string- and lice length.

# Average and the median egg string length of lice from farmed and wild origin.

#Average
data %>%
  select(Av.egg.string.L, origin) %>%
  group_by(origin) %>%
  summarise(mean(Av.egg.string.L))
```

```
# Median
data %>%
  select(Av.egg.string.L, origin) %>%
  group_by(origin) %>%
  summarise(median(Av.egg.string.L))

#  Average and the median lice length of lice from farmed and wild origin.

# Average
data %>%
  select(Lice.L, origin) %>%
  group_by(origin) %>%
  summarise(mean(Lice.L))

# Median
data %>%
  select(Lice.L, origin) %>%
  group_by(origin) %>%
  summarise(median(Lice.L))

# Average and the median number of eggs of lice from farmed and wild origin.

#Average
data %>%
  select(Total.eggs, origin) %>%
  group_by(origin) %>%
  summarise(mean(Total.eggs))

# Median
data %>%
  select(Total.eggs, origin) %>%
  group_by(origin) %>%
  summarise(median(Total.eggs))


# 2 Body length and number of eggs of lice from different hosts

plot2 <-data %>% select(Host, Total.eggs,Lice.L) %>%
  rename("b) Average body length (mm)" = Lice.L, "a) Average number of eggs"=Total.eggs) %>%
  pivot_longer(cols = c("a) Average number of eggs", "b) Average body length (mm)"), names_to = "variable",
values_to = 'value') %>%
  ggplot(aes(x=Host, y=value, fill=Host)) +
  geom_violin(trim = FALSE) +
  scale_fill_brewer(palette="Blues") +
  #geom_jitter(shape=16, position = position_jitter(0.2))+
  geom_boxplot(width=0.05, fill='white') +
  theme_bw() +
  coord_flip() +
  theme(text=element_text(size=20,  family="Times New Roman"), plot.title = element_text(size=20)) +
  labs(x="Species", y="") +
    facet_wrap(~variable, scales = "free_x")

plot2

ggsave("1plot2cordflip.png", plot2, height = 6, width = 11)



# IQR calculations of  the number of eggs and body length of different hosts
```

```
data1<-data %>% select(Host, Total.eggs,Lice.L) %>%
  rename("b) Average body length (mm)" = Lice.L, "a) Average number of eggs"=Total.eggs) %>%
  pivot_longer(cols = c("a) Average number of eggs", "b) Average body length (mm)"), names_to = "variable",
values_to = 'value')

# IQR of the average number of eggs
summary(data1 %>% filter(Host=="Arctic char" & variable=="a) Average number of eggs"))
summary(data1 %>% filter(Host=="Atlantic salmon" & variable=="a) Average number of eggs"))
summary(data1 %>% filter(Host=="Garfish" & variable=="a) Average number of eggs"))
summary(data1 %>% filter(Host=="Lumpfish" & variable=="a) Average number of eggs"))
summary(data1 %>% filter(Host=="Trout" & variable=="a) Average number of eggs"))

# IQR of the average body length
summary(data1 %>% filter(Host=="Arctic char" & variable=="b) Average body length (mm)"))
summary(data1 %>% filter(Host=="Atlantic salmon" & variable=="b) Average body length (mm)"))
summary(data1 %>% filter(Host=="Garfish" & variable=="b) Average body length (mm)"))
summary(data1 %>% filter(Host=="Lumpfish" & variable=="b) Average body length (mm)"))
summary(data1 %>% filter(Host=="Trout" & variable=="b) Average body length (mm)"))


# 4 Boxplot of the number of C. elongatus eggs of lice with wild and farmed origin.

plot4 <- data%>% group_by(origin) %>%
  ggplot(aes(x=origin, y=Total.eggs, fill=origin)) +
  scale_fill_manual(values = c("steelblue4", "lightskyblue2")) +
  geom_boxplot(width=0.3) +
  theme_minimal() +
  theme(text=element_text(size=20,  family="Times New Roman"), plot.title = element_text(size=20)) +
  labs(x="Origin", y="Number of eggs", fill='Origin') +
  scale_y_continuous(limits = c(0, 225))

plot4

ggsave("plot4box.png", plot4, height = 4, width = 5)

# IQR calculations of the number of eggs from lice with farmed and wild origin.

data2 <-data %>% select(origin, Total.eggs)
summary(data2 %>% filter(origin=="Farmed"))
summary(data2 %>% filter(origin=="Wild"))


# 5 Bar chart of the mean abundance of sea lice from the sampling days.

data1 <- read_excel("liceposition.xlsx")

data_summary <- data1 %>%
  group_by(area, stage) %>%
  summarise(mean = mean(number),
            sd = sd(number),
            n = n(),
            SE = sd(number)/sqrt(n())) %>%
mutate(stage= case_when(stage=="M"~"S2", stage=="F"~"S1"))

plot5 <- ggplot(data_summary, mapping=aes(x=area, y=mean, fill=stage)) +
  geom_col(colour="black", position = "dodge") +
  theme_minimal() +
  theme(text=element_text(size=20,  family="Times New Roman"), plot.title = element_text(size=20)) +
  labs(x="Position", y="Mean abundance of sea lice", fill="Stage")  +
```

```
  geom_errorbar(aes(ymin = mean, ymax = mean + sd), width=0.2, position=position_dodge(0.9)) +
  scale_fill_manual(values = c("steelblue4", "lightskyblue2"))

plot5

ggsave("plot5.png", plot5, height = 4, width = 7)


# 6 Bar chart of the nauplius duration of C. elongatus from N1 – N2 and from N2 to C.

df_10 <- read.csv("~/Desktop/R/molting frequence.csv", sep = ";", dec=",") %>%
  rename("M1"="H...N2", "M2" = "N2...C") %>%
  as_tibble() %>%
  filter(M1>0) %>%
  arrange(M1, M2) %>%
  mutate(Lice=1:n()) %>%
  pivot_longer(cols=c("M1","M2"), names_to = "Molting", values_to = "Days")

plot6 <- ggplot(df_10, aes(y = Days, x = Lice, fill = Molting)) +
  geom_col(width=0.7, colour="black", position = position_stack(reverse = TRUE), show.legend = TRUE) +
  theme_minimal() +
  theme(text=element_text(size=20,  family="Times New Roman"), plot.title = element_text(size=25)) +
  scale_fill_manual(values = c("lightskyblue2","steelblue4"))

plot6

ggsave("plot6.png", plot6, height = 4, width = 7)


#Map and bubbleplot R script

library(rgdal)
library(ggmap)
library(tidyverse)
library(RColorBrewer)
library(ggrepel)
library(extrafont)

mapsize <- c(left = 4.3,
             bottom = 57.7,
             right = 31.5,
             top = 71.3)

silje_stamen <- get_stamenmap(bbox = mapsize, zoom = 5, silje <- ggmap(silje_stamen)

# Importing the data

setwd("~/Desktop/R")

coordinates <- read.csv("Coordinates1.csv", header = TRUE, sep = ";", dec = ".") %>%
  mutate(origin= case_when( Posisjon=="M" ~ "Farmed", TRUE  ~ "Wild"))

ggmap(silje_stamen) +
  geom_point(data=coordinates, mapping=aes(x=Lon, y=Lat, colour=origin), size=3) +
  scale_color_manual(values=c("red", "black"))
  geom_text(data=coordinates, mapping=aes(label=Posisjon))

ggsave("kart.png", 'kart', height = 10, width = 5)
```

Statistica output:

Table J1. *Mann-Whitney U Test (w/ continuity correction) By variable origin (Farmed-Wild) Results highlighted in red are significant at p < ,05000.*

Variable	Rank Sum Gr. 1	Rank Sum Gr. 2	U	Z	p-value	Z adjusted	p-value	Valid N Gr. 1	Valid N Gr. 2
Eggs Tot	34786.00	6830.00	3509.00	7.670074	0.0000	7.670763	0.0000	207	81

Table J2. *Kruskal-Wallis ANOVA by Ranks; Eggs Tot. Independent (grouping) variable: Region Kruskal-Wallis test: H (2, N= 207) =13,52942 p =,0012.*

Depend: Eggs Tot	Code	Valid N	Sum of Ranks	Mean Rank
1	1	22	3144.00	142,9091
2	2	6	852.00	142,000
3	3	179	17532.00	97,9441

Table J3. *Post-Hoc MC (Multiple Comparisons) p values (2-tailed); Eggs Tot Independent (grouping) variable: Region Kruskal-Wallis test: H (2, N= 207) =13,52942 p =,0012.*

Depend: Eggs Tot	1 = North R:142.91	2 = Mid R:142.00	3 = South R: 97.944
1 North		1.0000	0.002675
2 Central	1.0000		0.229123
3 South	0.002675	0.229123	

Table J4. *Mann-Whitney U Test (w/ continuity correction) By variable origin (Farmed-Wild). Results highlighted in red are significant at p < ,05000.*

Variable	Rank Sum Gr. 1	Rank Sum Gr. 1	U	Z	p-vaule	Z adjusted	p-value	Valid N Gr. 1	Valid N Gr. 1
Eggs Tot	27333,50	6596,50	3275,50	7,075784	0,0000	7,076543	0,0000	179	81

Table J5. *T-test of the total number of eggs (Eggs Tot) and the lice length (Lice L). Results highlighted in red are significant at p < ,05000.*

X & Y	Region	Origin	Mean	Std. Dv.	R(X,Y)	r^2	t	p	N
Eggs Tot	North	Wild	154,5455	36,97454					
Lice L	North	Wild	5,87230	0,378530	0,245488	0,060264	1,132511	0,270815	22
Eggs Tot	Central	Wild	149,000	25,93839					
Lice L	Central	Wild	5,92000	0,604520	0,281885	0,079459	0,587597	0,588372	6
Eggs Tot	South	Wild	126,5587	34,48531					
Lice L	South	Wild	5,77510	0,360380	0,151107	0,022833	2,033694	0,043475	179
Eggs Tot	South	Farmed	95,09877	23,08604					

Lice L	South	Farmed	5,43370	0,523460	0,23772	0,056511	2,175255	0,032601	81

Table J6. *Kruskal-Wallis ANOVA by Ranks; Eggs Tot. Independent (grouping) variable: Host Kruskal-Wallis test: H (4, N= 285) =72.5310 p =,0000.1= Arctic char, 2= Atlantic salmon, 3=Garfish, 4= Lumpfish, 5= Sea trout.*

Depend: Egg Tot	Code	Valid N	Sum of Ranks	Mean Rank
1	1	9	2073.5	230.388889
2	2	82	6840	83.4146341
3	3	144	22362	155.291667
4	4	28	5484.5	195.875
5	5	22	3995	181.590909

Table J7. *Post-Hoc MC (Multiple Comparisons) p values (2-tailed); Eggs Tot Independent (grouping) variable: Host specie Kruskal-Wallis test: H (4, N= 285) =72.5310 p =,0000.1.*

Depend: Eggs tot	Atlantic Charr: 1 R: 230.389	Atlantic Salmon: 2 R: 83.415	Garfish: 3 R: 155.292	Lumpfish: 4 R: 195.875	Sea trout: 5 R: 181,591
Atlantic Charr: 1		0.000	0.0080	0.2744	0.1345
Atlantic Salmon: 2	0.0000		0.000	0.000	0.000
Garfish: 3	0.0080	0.000		0.0171	0.1633
Lumpfish: 4	0.2744	0.000	0.0171		0.5429
Sea trout: 5	0.1345	0.000	0.1633	0.5429	

Table J8. *Kruskal-Wallis ANOVA by Ranks; Eggs Tot. Independent (grouping) variable: Host Kruskal-Wallis test: H (4, N= 285) =5,1223 p =,2749.1= Arctic char, 2= Atlantic salmon, 3=Garfish, 4= Lumpfish, 5= Sea trout.*

Depend: Lice length	Code	Valid N	Sum of Ranks	Mean Rank
1	1	9	1374	152.666667
2	2	82	7998	97.5365854
3	3	144	24484	170.027778
4	4	28	4360.5	155.732143
5	5	22	2538.5	115.386364

7.9 APPENDIX I – Dataset

Table 1I. *Dataset used for estimations of the fecundity of 287 C. elongatus females. The locality where the C. elongatus host was captured, the date, previously fish host, number of eggs in the left- and right egg string, the total number of eggs, the body length of the lice (mm), left- and right egg string length (mm), combined average egg string length for left and right egg string (mm), number of eggs pr mm of the left- and right egg string, and the average number of eggs pr mm egg string for both left and right egg string. In addition, C. elongatus collected from host species with the farmed origin is marked (*).*

The locality of fish host	Date of capture	Previous host of C. elongatus	Number of eggs in egg string (left)	Number of eggs in egg string (right)	Total number of eggs	Lice length (mm)	Egg string length (mm) (left)	Egg string length (mm) (right)	Av. egg string length (mm) (left, right)	Number of eggs pr mm egg string (left)	Number of eggs pr mm egg string (right)	Av. number of eggs pr mm egg string (left, right)
Bugøyfjord	20.07.19	Sea trout	87	20	107	5,88	7,48	3,27	5,38	11,63	6,120	9,95
Jarfjorden	31.07.19	Garfish	69	68	137	6,17	4,23	4,18	4,21	16,32	16,26	16,29
Bugøyfjorden	29.07.19	Unknown	63	55	118	6,21	4,16	3,83	3,99	15,16	14,35	14,77
Namsenfjorden	09.06.19	Garfish	67	77	144	6,70	6,60	7,65	7,13	10,15	10,06	10,10
Ullsfjorden	17.07.19	Arctic char	73	78	151	6,31	4,82	4,99	4,90	15,16	15,64	15,40
Reisafjorden	17.07.19	Sea trout	42	41	83	5,47	2,77	2,68	2,73	15,18	15,28	15,23
Reisafjorden	17.07.19	Arctic char	56	55	111	5,83	3,43	3,46	3,44	16,35	15,89	16,12
Reisafjorden	17.07.19	Arctic char	99	97	196	6,13	6,36	6,20	6,28	15,57	15,66	15,62
Ullsfjorden	15.07.19	Sea trout	87	92	179	5,81	5,27	5,32	5,29	16,51	17,31	16,91
Ullsfjorden	18.07.19	Sea trout	92	91	183	5,53	5,70	5,41	5,55	16,15	16,82	16,48
Ullsfjorden	15.07.19	Lumpfish	103	101	204	6,23	6,01	5,85	5,93	17,13	17,27	17,20
Ullsfjorden	15.07.19	Lumpfish	98	96	194	6,10	5,96	5,87	5,91	16,44	16,37	16,40
Ullsfjorden	15.07.19	Lumpfish	85	103	188	6,17	3,77	3,85	3,81	22,55	26,79	24,69
Ullsfjorden	15.07.19	Sea trout	88	87	175	6,12	4,19	4,17	4,18	21,02	20,84	20,93
Øksfjorden	20s.07.19	Lumpfish	70	71	141	5,53	6,52	6,44	6,48	10,73	11,03	10,88
Ullsfjorden	15.07.19	Arctic char	94	87	181	6,36	5,33	5,42	5,38	17,62	16,05	16,83
Ullsfjorden	15.07.19	Arctic char	57	53	110	6,23	6,07	6,08	6,07	9,40	8,72	9,06

Reisafjorden	15.07.19	Arctic char	89	87	176	5,19	5,06	5,31	5,18	17,59	16,40	16,98
Reisafjorden	15.07.19	Arctic char	90	90	180	5,30	5,24	4,98	5,11	17,18	18,08	17,62
Altafjorden	20.07.19	Arctic char	91	93	184	5,77	4,76	2,35	3,55	19,12	39,58	25,88
Porsangerfjorden	22.07.19	Sea trout	35	55	90	5,14	5,83	5,81	5,82	6,01	9,47	7,74
Porsangerfjorden	21.07.19	Sea trout	78	71	149	5,62	4,68	4,62	4,65	16,67	15,36	16,02
Blindalsfjorden	28.06.19	Sea trout	84	83	167	5,95	7,73	7,69	7,71	10,87	10,79	10,83
Blindalsfjorden	28.06.19	Sea trout	47	53	100	5,47	6,76	6,22	6,49	6,95	8,52	7,70
Blindalsfjorden	28.06.19	Sea trout	87	84	171	5,95	8,69	8,14	8,41	10,01	10,32	10,16
Blindalsfjorden	28.06.19	Sea trout	81	79	160	6,41	6,74	6,75	6,75	12,01	11,70	11,86
Blindalsfjorden	28.06.19	Sea trout	76	76	152	5,04	5,46	5,40	5,43	13,93	14,07	14,00
Nordfjorden	15.05.19	Unknown	75	77	152	4,77	6,27	6,00	6,13	11,97	12,83	12,39
Nordfjorden	15.05.19	Unknown	96	92	188	5,35	6,11	6,50	6,30	15,72	14,16	14,92
Nordfjorden	02.07.19	Sea trout	112	113	225	5,41	7,29	7,50	7,39	15,36	15,07	15,22
Nordfjorden	02.07.19	Sea trout	89	96	185	6,14	6,13	6,07	6,10	14,51	15,82	15,17
Nordfjorden	02.07.19	Sea trout	55	52	107	5,64	3,45	3,33	3,39	15,96	15,60	15,78
Nordfjorden	02.07.19	Sea trout	100	101	201	5,51	4,87	4,70	4,79	20,53	21,49	21,00
Nordfjorden	02.07.19	Sea trout	46	49	95	5,82	4,18	4,15	4,17	11,01	11,80	11,40
Nordfjorden	02.07.19	Sea trout	109	108	217	5,57	4,99	4,99	4,99	21,83	21,63	21,73
Altafjorden	20.07.19	Arctic char	84	79	163	6,09	5,51	5,48	5,50	15,24	14,42	14,83
Boknafjorden	09.05.19	Garfish	57	62	119	6,15	6,28	6,11	6,19	9,07	10,15	9,61
Boknafjorden	09.05.19	Garfish	55	50	105	5,81	5,78	6,25	6,01	9,51	8,01	8,73
Boknafjorden	09.05.19	Garfish	31	39	70	4,81	4,83	4,76	4,79	6,42	8,20	7,30
Boknafjorden	09.05.19	Garfish	88	88	176	4,75	5,92	5,72	5,82	14,87	15,39	15,12
Boknafjorden	09.05.19	Garfish	69	67	136	6,34	4,98	4,88	4,93	13,86	13,74	13,80
Boknafjorden	09.05.19	Garfish	44	44	88	6,35	5,61	5,51	5,56	7,84	7,98	7,91
Boknafjorden	09.05.19	Garfish	60	63	123	6,16	5,33	5,38	5,35	11,25	11,72	11,49
Boknafjorden	09.05.19	Garfish	63	60	123	6,02	3,22	3,77	3,50	19,55	15,92	17,59
Boknafjorden	09.05.19	Garfish	32	35	67	5,83	4,82	4,57	4,70	6,63	7,66	7,13

Boknafjorden	09.05.19	Garfish	77	77	154	6,38	4,85	4,81	4,83	15,86	16,02	15,94
Boknafjorden	09.05.19	Garfish	59	67	126	6,47	2,87	2,93	2,90	20,54	22,88	21,72
Boknafjorden	09.05.19	Garfish	44	37	81	6,26	5,34	5,21	5,28	8,24	7,10	7,68
Boknafjorden	09.05.19	Garfish	52	63	115	6,06	5,37	5,21	5,29	9,69	12,09	10,87
Boknafjorden	09.05.19	Garfish	52	52	104	5,84	4,48	4,42	4,45	11,60	11,77	11,69
Boknafjorden	09.05.19	Garfish	59	59	118	5,30	5,11	5,05	5,08	11,55	11,69	11,62
Boknafjorden	09.05.19	Garfish	84	83	167	5,98	5,60	5,94	5,77	15,00	13,98	14,48
Boknafjorden	09.05.19	Garfish	73	71	144	6,03	3,36	3,44	3,40	21,73	20,65	21,18
Boknafjorden	20.05.19	Garfish	55	60	115	5,94	2,92	3,07	2,99	18,86	19,54	19,21
Boknafjorden	20.05.19	Garfish	64	63	127	5,62	6,05	5,64	5,84	10,58	11,17	10,87
Boknafjorden	20.05.19	Garfish	55	56	111	5,74	3,39	3,89	3,64	16,20	14,41	15,24
Boknafjorden	20.05.19	Garfish	42	42	84	6,19	6,28	3,97	5,13	6,68	10,59	8,19
Boknafjorden	20.05.19	Garfish	81	81	162	5,83	6,05	5,64	5,85	13,39	14,36	13,86
Boknafjorden	20.05.19	Garfish	72	62	134	5,92	5,07	5,02	5,05	14,20	12,34	13,27
Boknafjorden	20.05.19	Garfish	56	56	112	5,80	4,95	5,83	5,39	11,32	9,61	10,39
Boknafjorden	20.05.19	Garfish	88	95	183	6,09	3,65	3,68	3,67	24,11	25,80	24,96
Boknafjorden	20.05.19	Garfish	59	59	118	6,09	4,34	6,31	5,33	13,59	9,35	11,08
Boknafjorden	20.05.19	Garfish	88	85	173	5,99	5,22	5,75	5,49	16,85	14,77	15,76
Boknafjorden	20.05.19	Garfish	66	72	138	5,99	4,09	3,82	3,96	16,15	18,82	17,44
Boknafjorden	20.05.19	Garfish	74	64	138	6,05	5,38	4,52	4,95	13,75	14,15	13,93
Boknafjorden	20.05.19	Garfish	78	78	156	5,97	4,74	4,83	4,78	16,45	16,16	16,31
Boknafjorden	20.05.19	Garfish	76	79	155	5,75	6,89	6,35	6,62	11,02	12,45	11,71
Boknafjorden	20.05.19	Garfish	78	78	156	6,10	4,99	4,34	4,67	15,62	17,98	16,72
Boknafjorden	20.05.19	Garfish	50	50	100	5,84	5,87	5,52	5,70	8,51	9,06	8,78
Boknafjorden	20.05.19	Garfish	21	20	41	5,89	5,06	5,05	5,05	4,15	3,96	4,06
Boknafjorden	20.05.19	Garfish	89	89	178	5,82	4,66	4,69	4,68	19,10	18,97	19,04
Boknafjorden	20.05.19	Garfish	67	67	134	6,20	6,09	6,22	6,15	11,00	10,78	10,89
Boknafjorden	20.05.19	Garfish	55	65	120	6,26	5,60	5,42	5,51	9,83	11,99	10,89

Boknafjorden	20.05.19	Garfish	67	67	134	5,44	7,09	6,96	7,02	9,45	9,63	9,54
Boknafjorden	20.05.19	Garfish	38	38	76	5,92	5,73	5,91	5,82	6,64	6,43	6,53
Boknafjorden	20.05.19	Garfish	74	64	138	5,67	6,31	6,70	6,51	11,72	9,55	10,60
Boknafjorden	20.05.19	Garfish	64	68	132	5,92	6,46	6,17	6,32	9,91	11,02	10,45
Boknafjorden	20.05.19	Garfish	40	34	74	5,80	7,49	7,34	7,41	5,34	4,63	4,99
Boknafjorden	20.05.19	Garfish	78	78	156	5,74	3,58	4,49	4,04	21,78	17,37	19,32
Boknafjorden	20.05.19	Garfish	66	66	132	6,00	7,43	7,30	7,36	8,88	9,05	8,96
Boknafjorden	02.05.19	Garfish	84	91	175	5,79	6,24	6,23	6,24	13,47	14,60	14,03
Boknafjorden	02.05.19	Garfish	56	64	120	5,66	6,37	6,51	6,44	8,80	9,83	9,32
Boknafjorden	02.05.19	Garfish	43	43	86	5,55	3,48	3,51	3,49	12,36	12,25	12,30
Boknafjorden	02.05.19	Garfish	52	49	101	5,70	7,02	7,21	7,12	7,41	6,79	7,10
Boknafjorden	02.05.19	Garfish	66	68	134	5,47	3,34	3,24	3,29	19,76	20,96	20,35
Boknafjorden	02.05.19	Garfish	55	55	110	5,97	6,23	6,49	6,36	8,83	8,48	8,65
Boknafjorden	02.05.19	Garfish	66	68	134	5,46	5,46	5,63	5,54	12,08	12,09	12,08
Boknafjorden	08.05.19	Lumpfish	47	45	92	5,95	5,40	5,48	5,44	8,71	8,21	8,46
Boknafjorden	08.05.19	Lumpfish	74	78	152	5,51	3,73	3,77	3,75	19,86	20,70	20,28
Boknafjorden	08.05.19	Lumpfish	85	85	170	6,11	5,05	4,69	4,87	16,82	18,12	17,45
Boknafjorden	09.05.19	Garfish	45	43	88	5,90	5,13	5,37	5,25	8,78	8,01	8,39
Boknafjorden	09.05.19	Garfish	52	49	101	5,98	5,63	4,95	5,29	9,23	9,90	9,54
Boknafjorden	07.05.19	Lumpfish	45	50	95	5,60	5,56	6,62	6,09	8,09	7,56	7,80
Boknafjorden	07.05.19	Lumpfish	84	83	167	5,97	3,37	3,53	3,45	24,92	23,52	24,21
Boknafjorden	30.04.19	Garfish	34	33	67	5,87	2,95	3,31	3,13	11,53	9,96	10,70
Boknafjorden	30.04.19	Garfish	35	34	69	5,54	3,57	3,43	3,50	9,80	9,90	9,85
Boknafjorden	30.04.19	Garfish	38	78	116	6,41	3,31	6,64	4,97	11,48	11,75	11,66
Boknafjorden	30.04.19	Garfish	79	84	163	6,01	6,64	6,73	6,69	11,90	12,47	12,19
Boknafjorden	30.04.19	Garfish	54	47	101	5,65	4,62	4,96	4,79	11,70	9,47	10,55
Boknafjorden	30.04.19	Garfish	64	65	129	5,79	6,54	6,42	6,48	9,79	10,12	9,96
Boknafjorden	09.05.19	Lumpfish	59	58	117	5,30	3,87	3,83	3,85	15,23	15,15	15,19

Boknafjorden	09.05.19	Lumpfish	106	102	208	6,31	6,17	6,09	6,13	17,17	16,75	16,96
Boknafjorden	08.05.19	Garfish	63	57	120	6,21	6,03	5,62	5,83	10,45	10,14	10,30
Boknafjorden	08.05.19	Garfish	69	67	136	5,84	5,71	5,52	5,62	12,08	12,13	12,11
Boknafjorden	05.05.19	Garfish	61	64	125	5,71	5,65	5,58	5,62	10,79	11,47	11,13
Boknafjorden	05.05.19	Garfish	59	58	117	5,86	4,61	4,84	4,73	12,79	11,98	12,38
Boknafjorden	05.05.19	Garfish	51	56	107	5,50	4,77	4,66	4,71	10,70	12,01	11,35
Boknafjorden	06.05.19	Lumpfish	36	35	71	5,36	2,34	2,25	2,30	15,41	15,52	15,47
Boknafjorden	06.05.19	Lumpfish	66	65	131	5,45	4,02	4,04	4,03	16,43	16,08	16,25
Boknafjorden	13.05.19	Garfish	52	61	113	5,73	4,65	5,46	5,06	11,18	11,18	11,18
Boknafjorden	13.05.19	Garfish	31	31	62	5,65	3,19	2,13	2,66	9,73	14,58	11,67
Boknafjorden	13.05.19	Lumpfish	58	64	122	4,74	3,43	3,61	3,52	16,93	17,74	17,34
Boknafjorden	06.05.19	Lumpfish	107	95	202	6,01	5,36	5,63	5,50	19,95	16,88	18,38
Boknafjorden	29.04.19	Lumpfish	70	70	140	5,74	4,77	4,81	4,79	14,69	14,57	14,63
Boknafjorden	04.05.19	Garfish	58	49	107	6,05	6,61	6,17	6,39	8,78	7,94	8,37
Boknafjorden	04.05.19	Garfish	62	56	118	6,31	6,33	6,38	6,35	9,80	8,78	9,29
Boknafjorden	05.05.19	Lumpfish	72	80	152	4,88	4,04	4,15	4,10	17,83	19,26	18,55
Boknafjorden	09.05.19	Garfish	73	67	140	6,01	6,04	5,53	5,78	12,10	12,11	12,10
Boknafjorden	09.05.19	Garfish	81	81	162	5,96	7,12	6,73	6,92	11,38	12,04	11,70
Boknafjorden	09.05.19	Garfish	65	62	127	6,16	5,58	5,73	5,65	11,64	10,83	11,23
Boknafjorden	09.05.19	Garfish	50	50	100	5,68	5,70	5,37	5,53	8,77	9,32	9,04
Boknafjorden	09.05.19	Garfish	64	68	132	5,62	6,42	6,39	6,40	9,98	10,65	10,31
Boknafjorden	02.05.19	Garfish	58	59	117	6,05	5,68	5,62	5,65	10,22	10,50	10,36
Boknafjorden	02.05.19	Garfish	86	84	170	5,81	7,13	6,95	7,04	12,05	12,09	12,07
Boknafjorden	02.05.19	Garfish	75	75	150	6,21	6,18	6,07	6,12	12,15	12,35	12,25
Boknafjorden	10.05.19	Garfish	59	54	113	6,20	5,57	5,54	5,55	10,60	9,75	10,18
Boknafjorden	10.05.19	Garfish	52	53	105	6,20	5,47	5,41	5,44	9,50	9,79	9,64
Boknafjorden	10.05.19	Garfish	59	58	117	6,13	5,95	5,99	5,97	9,91	9,68	9,80
Boknafjorden	10.05.19	Garfish	58	55	113	5,66	5,36	5,13	5,24	10,83	10,73	10,78

Boknafjorden	10.05.19	Garfish	38	45	83	5,48	4,41	3,73	4,07	8,62	12,06	10,19
Boknafjorden	10.05.19	Garfish	58	54	112	6,16	5,76	5,31	5,53	10,07	10,18	10,12
Boknafjorden	10.05.19	Garfish	28	42	70	5,53	2,63	3,68	3,15	10,64	11,43	11,10
Boknafjorden	08.05.19	Garfish	65	68	133	5,92	5,93	5,87	5,90	10,97	11,59	11,28
Boknafjorden	10.05.19	Garfish	51	55	106	5,49	5,08	4,84	4,96	10,04	11,36	10,69
Boknafjorden	10.05.19	Garfish	68	66	134	5,97	7,25	6,75	7,00	9,37	9,78	9,57
Boknafjorden	10.05.19	Garfish	89	83	172	5,87	7,48	7,91	7,69	11,89	10,50	11,18
Boknafjorden	10.05.19	Garfish	66	63	129	5,93	6,43	6,40	6,41	10,27	9,85	10,06
Boknafjorden	10.05.19	Garfish	63	65	128	5,83	7,96	7,79	7,87	7,91	8,35	8,13
Boknafjorden	10.05.19	Lumpfish	76	75	151	5,57	4,92	4,74	4,83	15,45	15,82	15,63
Boknafjorden	04.05.19	Lumpfish	84	81	165	5,86	8,04	7,77	7,90	10,45	10,43	10,44
Boknafjorden	04.05.19	Lumpfish	73	82	155	5,90	6,68	7,17	6,92	10,93	11,44	11,19
Boknafjorden	04.05.19	Lumpfish	34	37	71	5,98	3,49	3,31	3,40	9,75	11,17	10,44
Boknafjorden	04.05.19	Lumpfish	70	72	142	6,34	7,19	7,63	7,41	9,73	9,43	9,58
Boknafjorden	11.05.19	Garfish	52	56	108	5,25	4,71	5,05	4,88	11,05	11,08	11,07
Boknafjorden	11.05.19	Garfish	89	90	179	5,93	7,95	7,49	7,72	11,20	12,02	11,60
Boknafjorden	11.05.19	Garfish	49	48	97	5,73	3,97	3,90	3,93	12,35	12,31	12,33
Boknafjorden	11.05.19	Garfish	71	61	132	6,06	6,22	6,33	6,27	11,41	9,64	10,52
Boknafjorden	11.05.19	Garfish	33	41	74	5,97	3,05	3,30	3,17	10,83	12,44	11,67
Boknafjorden	11.05.19	Garfish	73	69	142	5,70	6,91	6,92	6,92	10,57	9,97	10,27
Boknafjorden	11.05.19	Garfish	62	30	92	5,91	5,52	3,72	4,62	11,23	8,06	9,95
Boknafjorden	11.05.19	Garfish	80	72	152	5,75	6,00	5,84	5,92	13,34	12,33	12,84
Boknafjorden	11.05.19	Garfish	71	79	150	5,48	6,41	6,88	6,65	11,07	11,48	11,28
Boknafjorden	11.05.19	Garfish	77	82	159	5,79	6,28	6,58	6,43	12,26	12,46	12,36
Boknafjorden	11.05.19	Garfish	46	49	95	6,05	5,02	5,39	5,21	9,16	9,09	9,12
Boknafjorden	11.05.19	Garfish	77	66	143	5,78	6,88	6,51	6,69	11,20	10,14	10,68
Boknafjorden	11.05.19	Garfish	62	57	119	5,79	5,23	5,29	5,26	11,86	10,78	11,32
Boknafjorden	15.10.19	Garfish	52	47	99	5,54	6,48	5,30	5,89	8,02	8,87	8,40

Boknafjorden	15.05.19	Lumpfish	111	111	222	5,76	6,28	6,44	6,36	17,66	17,23	17,44
Boknafjorden	15.05.19	Lumpfish	30	27	57	4,55	2,08	1,91	2,00	14,39	14,17	14,28
Boknafjorden	15.05.19	Lumpfish	83	90	173	5,81	4,02	4,25	4,14	20,62	21,16	20,90
Boknafjorden	19.05.19	Garfish	68	72	140	6,26	5,80	5,60	5,70	11,72	12,86	12,28
Boknafjorden	19.05.19	Garfish	49	55	104	5,69	4,19	4,63	4,41	11,69	11,89	11,80
Boknafjorden	15.05.19	Lumpfish	96	102	198	6,55	5,72	6,26	5,99	16,79	16,30	16,53
Boknafjorden	15.05.19	Lumpfish	60	67	127	5,70	3,95	4,06	4,00	15,21	16,50	15,86
Boknafjorden	15.05.19	Lumpfish	50	54	104	5,83	2,71	2,89	2,80	18,43	18,66	18,55
Boknafjorden	15.05.19	Garfish	63	58	121	5,96	5,84	5,79	5,82	10,79	10,01	10,40
Boknafjorden	15.05.19	Garfish	67	54	121	5,98	5,24	5,39	5,32	12,78	10,02	11,38
Boknafjorden	15.05.19	Garfish	68	69	137	5,61	6,29	6,36	6,33	10,80	10,85	10,83
Boknafjorden	15.05.19	Garfish	60	68	128	6,38	5,89	5,21	5,55	10,18	13,06	11,53
Boknafjorden	15.05.19	Garfish	68	69	137	5,56	5,74	5,89	5,81	11,84	11,72	11,78
Boknafjorden	15.05.19	Garfish	53	41	94	5,61	5,20	3,89	4,54	10,19	10,55	10,35
Boknafjorden	15.05.19	Garfish	61	65	126	5,81	6,51	6,06	6,29	9,37	10,73	10,02
Boknafjorden	15.05.19	Garfish	81	76	157	5,94	7,43	7,85	7,64	10,90	9,68	10,27
Boknafjorden	15.05.19	Garfish	55	66	121	5,46	6,05	6,43	6,24	9,10	10,26	9,70
Boknafjorden	19.05.19	Garfish	78	55	133	5,88	7,87	5,20	6,53	9,91	10,58	10,18
Boknafjorden	19.05.19	Garfish	64	63	127	6,12	5,03	4,78	4,91	12,72	13,18	12,95
Boknafjorden	19.05.19	Garfish	46	47	93	5,39	4,93	5,12	5,02	9,33	9,19	9,26
Boknafjorden	19.05.19	Garfish	78	80	158	5,78	7,02	7,17	7,09	11,11	11,16	11,13
Boknafjorden	19.05.19	Garfish	53	44	97	6,18	5,36	4,86	5,11	9,89	9,05	9,49
Boknafjorden	19.05.19	Garfish	71	61	132	6,00	7,04	6,02	6,53	10,08	10,13	10,11
Boknafjorden	19.05.19	Garfish	28	26	54	5,63	1,71	1,55	1,63	16,36	16,74	16,54
Boknafjorden	19.05.19	Garfish	46	45	91	5,39	4,02	4,30	4,16	11,45	10,46	10,94
Boknafjorden	19.05.19	Garfish	47	47	94	5,91	3,98	4,00	3,99	11,81	11,76	11,79
Boknafjorden	19.05.19	Garfish	70	67	137	5,62	6,65	6,47	6,56	10,52	10,36	10,44
Boknafjorden	16.05.19	Garfish	78	88	166	5,58	6,73	6,75	6,74	11,58	13,03	12,31

Boknafjorden	16.05.19	Garfish	51	50	101	5,74	5,87	5,30	5,59	8,69	9,43	9,04
Boknafjorden	16.05.19	Garfish	46	41	87	5,55	4,65	3,96	4,31	9,88	10,35	10,10
Boknafjorden	16.05.19	Garfish	64	66	130	5,84	5,91	6,22	6,07	10,83	10,61	10,71
Boknafjorden	16.05.19	Garfish	75	73	148	5,82	6,43	6,60	6,52	11,66	11,06	11,36
Boknafjorden	16.05.19	Garfish	53	52	105	5,28	5,58	4,91	5,25	9,50	10,58	10,01
Boknafjorden	16.05.19	Garfish	82	81	163	5,49	6,32	6,46	6,39	12,98	12,54	12,76
Boknafjorden	12.05.19	Garfish	62	57	119	5,83	5,55	5,60	5,57	11,18	10,18	10,68
Boknafjorden	12.05.19	Garfish	55	62	117	5,60	4,80	5,63	5,22	11,46	11,00	11,21
Boknafjorden	12.05.19	Garfish	81	83	164	5,82	7,61	7,78	7,70	10,64	10,66	10,65
Boknafjorden	12.05.19	Garfish	46	53	99	5,32	3,56	4,37	3,96	12,93	12,13	12,49
Boknafjorden	16.05.19	Garfish	70	69	139	5,75	6,33	6,00	6,16	11,06	11,50	11,27
Boknafjorden	16.05.19	Garfish	69	73	142	5,36	5,95	5,92	5,93	11,60	12,33	11,97
Boknafjorden	16.05.19	Garfish	68	84	152	5,94	5,99	5,67	5,83	11,36	14,82	13,04
Boknafjorden	16.05.19	Garfish	76	78	154	5,95	6,11	6,03	6,07	12,44	12,94	12,69
Boknafjorden	16.05.19	Garfish	68	70	138	5,64	6,23	5,99	6,11	10,91	11,68	11,29
Boknafjorden	16.05.19	Garfish	43	50	93	5,48	4,22	4,59	4,41	10,19	10,89	10,55
Boknafjorden	16.05.19	Garfish	72	71	143	5,71	5,77	6,17	5,97	12,48	11,51	11,98
Sognefjorden	18.11.19	Atlantic salmon	31	42	73	4,99	1,99	2,64	2,32	15,55	15,91	15,75
Sognefjorden	16.11.19	Sea trout	48	46	94	5,00	3,51	3,01	3,26	13,69	15,26	14,42
Sognefjorden	16.11.19	Sea trout	39	40	79	4,81	4,98	5,03	5,00	7,83	7,95	7,89
Sognefjorden	18.11.19	Sea trout	40	43	83	4,94	2,64	2,69	2,66	15,17	15,99	15,58
Sognefjorden	18.11.19	Sea trout	81	82	163	4,83	4,11	4,11	4,11	19,71	19,94	19,83
Låva*	10.09.19	Atlantic salmon	45	44	89	4,70	3,11	3,12	3,12	14,45	14,12	14,28
Låva*	10.09.19	Atlantic salmon	51	50	101	4,87	3,07	3,10	3,09	16,59	16,11	16,35
Låva*	10.09.19	Atlantic salmon	54	56	110	5,52	3,38	3,46	3,42	15,97	16,19	16,08
Låva*	10.09.19	Atlantic salmon	51	47	98	6,08	3,01	3,03	3,02	16,94	15,52	16,23
Låva*	10.09.19	Atlantic salmon	48	50	98	5,26	3,10	3,29	3,20	15,48	15,19	15,33
Låva*	10.09.19	Atlantic salmon	38	38	76	4,53	2,72	2,68	2,70	13,96	14,19	14,07

Låva*	10.09.19	Atlantic salmon	57	69	126	5,75	4,13	4,27	4,20	13,81	16,15	15,00
Låva*	10.09.19	Atlantic salmon	47	47	94	5,38	3,44	3,49	3,46	13,67	13,48	13,57
Låva*	10.09.19	Atlantic salmon	47	47	94	5,02	3,46	3,50	3,48	13,59	13,43	13,51
Låva*	10.09.19	Atlantic salmon	50	50	100	4,15	2,85	2,90	2,88	17,52	17,24	17,38
Låva*	10.09.19	Atlantic salmon	35	35	70	4,16	3,07	3,06	3,07	11,38	11,43	11,41
Låva*	10.09.19	Atlantic salmon	38	43	81	5,18	3,23	3,25	3,24	11,75	13,22	12,49
Låva*	10.09.19	Atlantic salmon	43	43	86	6,21	3,18	3,15	3,17	13,52	13,65	13,59
Låva*	10.09.19	Atlantic salmon	45	46	91	5,64	2,84	2,98	2,91	15,83	15,43	15,62
Låva*	10.09.19	Atlantic salmon	26	48	74	5,47	2,71	3,52	3,11	9,60	13,65	11,89
Låva*	10.09.19	Atlantic salmon	48	47	95	5,28	3,14	3,10	3,12	15,30	15,15	15,22
Låva*	10.09.19	Atlantic salmon	59	59	118	5,46	3,92	3,96	3,94	15,05	14,92	14,98
Låva*	10.09.19	Atlantic salmon	61	61	122	5,95	3,77	3,84	3,81	16,17	15,88	16,02
Låva*	10.09.19	Atlantic salmon	40	40	80	4,87	2,88	2,91	2,90	13,87	13,74	13,81
Låva*	10.09.19	Atlantic salmon	60	62	122	5,93	4,37	4,24	4,31	13,72	14,62	14,16
Låva*	10.09.19	Atlantic salmon	60	58	118	6,07	3,60	3,65	3,62	16,69	15,88	16,28
Låva*	10.09.19	Atlantic salmon	41	39	80	5,92	3,32	3,38	3,35	12,37	11,54	11,95
Låva*	10.09.19	Atlantic salmon	53	56	109	5,34	3,55	3,73	3,64	14,91	15,01	14,96
Låva*	10.09.19	Atlantic salmon	75	80	155	6,40	5,14	5,13	5,13	14,59	15,59	15,09
Låva*	10.09.19	Atlantic salmon	49	49	98	5,10	3,32	3,25	3,28	14,77	15,09	14,93
Låva*	10.09.19	Atlantic salmon	48	49	97	5,57	3,48	3,40	3,44	13,81	14,41	14,11
Låva*	10.09.19	Atlantic salmon	51	51	102	5,34	3,34	3,35	3,35	15,26	15,22	15,24
Låva*	10.09.19	Atlantic salmon	63	36	99	5,44	4,20	3,09	3,64	14,99	11,67	13,58
Låva*	10.09.19	Atlantic salmon	23	25	48	5,24	1,84	1,94	1,89	12,47	12,90	12,69
Låva*	10.09.19	Atlantic salmon	24	26	50	5,17	2,42	2,56	2,49	9,94	10,18	10,06
Låva*	10.09.19	Atlantic salmon	57	57	114	5,66	3,40	3,30	3,35	16,77	17,28	17,02
Låva*	10.09.19	Atlantic salmon	47	47	94	4,67	3,27	3,29	3,28	14,38	14,29	14,34
Låva*	10.09.19	Atlantic salmon	59	19	78	5,80	4,23	2,34	3,28	13,96	8,12	11,88
Låva*	10.09.19	Atlantic salmon	51	50	101	5,33	3,83	3,99	3,91	13,32	12,55	12,93

Låva*	10.09.19	Atlantic salmon	46	46	92	5,56	3,29	3,27	3,28	14,00	14,07	14,04
Låva*	10.09.19	Atlantic salmon	53	53	106	5,10	3,62	3,61	3,61	14,66	14,69	14,67
Låva*	10.09.19	Atlantic salmon	56	55	111	5,16	4,08	4,28	4,18	13,72	12,85	13,28
Låva*	10.09.19	Atlantic salmon	34	32	66	6,11	3,72	3,61	3,67	9,13	8,87	9,00
Låva*	10.09.19	Atlantic salmon	49	49	98	5,12	3,42	3,53	3,48	14,31	13,88	14,09
Låva*	10.09.19	Atlantic salmon	60	53	113	6,37	6,74	6,37	6,55	8,91	8,32	8,62
Låva*	10.09.19	Atlantic salmon	53	48	101	5,81	3,88	3,64	3,76	13,65	13,17	13,42
Låva*	10.09.19	Atlantic salmon	30	19	49	5,39	2,81	2,33	2,57	10,67	8,17	9,54
Låva*	10.09.19	Atlantic salmon	37	45	82	6,14	3,87	4,07	3,97	9,55	11,05	10,32
Låva*	10.09.19	Atlantic salmon	59	54	113	5,52	3,99	3,77	3,88	14,79	14,34	14,57
Låva*	10.09.19	Atlantic salmon	54	53	107	5,13	3,69	3,47	3,58	14,64	15,29	14,95
Låva*	10.09.19	Atlantic salmon	49	52	101	6,12	3,57	3,57	3,57	13,71	14,57	14,14
Låva*	10.09.19	Atlantic salmon	52	53	105	5,71	3,74	3,91	3,82	13,92	13,56	13,73
Låva*	10.09.19	Atlantic salmon	54	53	107	5,31	3,93	3,77	3,85	13,74	14,05	13,89
Låva*	10.09.19	Atlantic salmon	41	39	80	4,85	2,94	3,03	2,98	13,97	12,87	13,41
Låva*	10.09.19	Atlantic salmon	74	67	141	6,56	5,31	4,90	5,11	13,93	13,66	13,80
Låva*	10.09.19	Atlantic salmon	54	50	104	5,67	5,95	5,67	5,81	9,07	8,82	8,95
Låva*	10.09.19	Atlantic salmon	57	55	112	6,32	6,20	6,03	6,11	9,20	9,12	9,16
Låva*	10.09.19	Atlantic salmon	50	43	93	5,99	3,76	3,45	3,60	13,29	12,48	12,90
Låva*	10.09.19	Atlantic salmon	47	49	96	5,19	3,45	3,55	3,50	13,61	13,80	13,71
Låva*	10.09.19	Atlantic salmon	41	39	80	4,90	3,45	3,33	3,39	11,88	11,72	11,80
Låva*	10.09.19	Atlantic salmon	60	59	119	5,24	4,19	4,24	4,21	14,33	13,92	14,12
Låva*	10.09.19	Atlantic salmon	48	45	93	5,56	3,27	3,20	3,24	14,66	14,05	14,36
Låva*	10.09.19	Atlantic salmon	56	56	112	5,42	3,94	3,96	3,95	14,21	14,15	14,18
Låva*	10.09.19	Atlantic salmon	28	28	56	6,26	2,98	3,00	2,99	9,39	9,33	9,36
Låva*	10.09.19	Atlantic salmon	27	27	54	5,14	2,71	2,76	2,73	9,95	9,80	9,87
Låva*	10.09.19	Atlantic salmon	51	50	101	4,87	3,50	3,52	3,51	14,59	14,19	14,39
Låva*	10.09.19	Atlantic salmon	14	14	28	5,91	2,14	2,16	2,15	6,56	6,50	6,53

Låva*	10.09.19	Atlantic salmon	63	59	122	5,22	4,14	3,98	4,06	15,21	14,83	15,02
Låva*	10.09.19	Atlantic salmon	38	48	86	5,55	3,39	3,95	3,67	11,20	12,16	11,72
Låva*	10.09.19	Atlantic salmon	43	43	86	5,51	3,33	3,27	3,30	12,92	13,15	13,03
Låva*	10.09.19	Atlantic salmon	67	70	137	5,37	4,46	4,72	4,59	15,04	14,84	14,94
Låva*	10.09.19	Atlantic salmon	49	52	101	4,82	3,96	3,98	3,97	12,38	13,06	12,72
Låva*	10.09.19	Atlantic salmon	62	63	125	5,55	3,99	4,08	4,03	15,52	15,46	15,49
Låva*	10.09.19	Atlantic salmon	58	68	126	6,38	4,22	4,38	4,30	13,73	15,51	14,64
Låva*	10.09.19	Atlantic salmon	50	43	93	5,16	3,11	3,19	3,15	16,06	13,47	14,75
Låva*	10.09.19	Atlantic salmon	39	38	77	5,32	2,84	2,83	2,83	13,76	13,44	13,60
Låva*	10.09.19	Atlantic salmon	48	54	102	5,61	3,79	3,72	3,75	12,66	14,54	13,59
Låva*	10.09.19	Atlantic salmon	58	61	119	5,78	3,89	4,02	3,96	14,92	15,16	15,04
Låva*	10.09.19	Atlantic salmon	40	44	84	5,15	3,18	3,18	3,18	12,60	13,83	13,22
Låva*	10.09.19	Atlantic salmon	28	28	56	5,34	2,31	2,31	2,31	12,14	12,12	12,13
Låva*	10.09.19	Atlantic salmon	48	48	96	4,05	3,37	3,29	3,33	14,23	14,61	14,42
Låva*	10.09.19	Atlantic salmon	15	26	41	4,83	1,86	2,77	2,31	8,06	9,40	8,86
Låva*	10.09.19	Atlantic salmon	48	49	97	5,55	3,43	3,41	3,42	14,00	14,39	14,19
Låva*	10.09.19	Atlantic salmon	47	48	95	5,76	3,60	3,71	3,65	13,05	12,95	13,00
Låva*	10.09.19	Atlantic salmon	29	30	59	5,60	2,76	2,80	2,78	10,52	10,71	10,62
Låva*	10.09.19	Atlantic salmon	56	57	113	4,69	3,67	3,71	3,69	15,27	15,38	15,33