Nematode infestation in flatfish in the inner and middle Oslofjord

by

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Abstract

Six flatfish species were collected by trawling at five locations in the inner and middle Oslofjord between February and December in 2013. In total 386 individuals were sampled. The purpose of this investigation was to identify the infection of *Hysterothylacium aduncum* and *Cucullanus heterochrous*. Only three species contained these parasites: American plaice (*Hippoglossoides platessoides*), European plaice (*Pleuronectes platessa*) and Witch flounder (*Glyptocephalus cynoglossus*). Also the condition, age distribution and weight-length relationship was measured on these fish species.

There was a geographical difference in the percentage of infected fish between the middle and inner part of the Oslofjord, as most nematodes were found in the middle part of the fjord. American plaice was the most abundant species and also the most infected specie. Two decades ago this species was not a known host, but during the last years this species has become an important host in the fjord.

Hysterothylacium aduncum infected all three fish species with large abundances in the middle Oslofjord. It seems that *H. aduncum* has been in an increasing trend over the last two decades. American plaice starts to mature at 2 and 3 years, and these age groups were most infected. Females were more infected than males. There was no correlation between the length of the nematodes and the condition of the host.

Cucullanus heterochrous was found in the inner Oslofjord, and this is only the second time this nematode has been reported in the inner part of the fjord, and both times it was found in European plaice and American plaice.

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1 Introduction

Nematodes are both free living and parasites in most warm and cold-blooded species and therefore constitute the most abundant species group on earth (Schmidt & Roberts 1985). Unlike many endoparasites, most nematodes have a complete digestive tract with mouth, oesophagus, intestine and anus (Berland 2006). They are thin, elongated, lacks segmentation and are bilaterally symmetrical (Schmidt & Roberts 1985; Berland 1973, 2006).

There are primarily five nematode species in the Norwegian coastal waters known to infect marine fish: *Anisakis simplex* (known as herring worm or whale worm), *Contracaecum osculatum*, *Pseudoterranova decipiens* (known as cod worm or seal worm), *Hysterothylacium aduncum* and *Cucullanus heterochrous* (Anderson 2000). Various fish species act as transport host or as final host. *Anisakis*, *Contracaecum* and *Pseudoterranova* become sexually mature in a warm-blooded animal, and use fish as a transport host until it reaches a marine mammal or piscivorous bird (final host). *Hysterothylacium* and *Cucullanus* become sexually mature in a “cold” intestine, and uses fish as the final host (Berland 1989).

People along the Norwegian coast use the word “kveis” for *Anisakis*, *Contracaecum* and *Pseudoterranova*, and are fully aware that these nematodes are very common in marine teleosts (Berland 1961, 1989). The larvae may be found in large abundances encysted on the liver, in the flesh and in the mesenteries (Berland 1961; Anderson 2000). Nematode larvae in marine fish can create a hygienic problem for the fishing industry and the removal of these parasites costs millions of Norwegian kroner each year.

There has been little research on the infection of flatfish in the inner Oslofjord. Most previous studies have been directed to the abundances in cod and seals in the outer Oslofjord by students at the University of Oslo. During their thesis project, Hansen & Malmstrøm (2006) found some flatfish species to be infected by nematodes in the inner and the outer Oslofjord, but did not find any infected fish in the middle Oslofjord. The fish nematode *H. aduncum* is especially common in the fjord (Andersen 1993; Raamat 2012).

The main purpose of my project was to investigate the infection of the nematodes in flatfishes from the middle and inner part of the Oslofjord. Five study areas were chosen, two from the middle Oslofjord (Drøbak and Tofteflaket) and three from the inner Oslofjord (Midtmeie, Hellvik and Gråøyrenna). At these sites there was a consistent catch of the following three species: American plaice (*Hippoglossoides platessoides*), European plaice (*Pleuronectes platessa*) and Witch flounder (*Glyptocephalus cynoglossus*). In addition to the infection by the nematodes *Hysterothylacium* and *Cucullanus,* this investigation also included the condition, age distribution and weight and length relationship for the flatfishes. These data are then discussed in relation to the increasing sea temperature which has been registered in the Oslofjord (Thaulow & Faafeng 2013). Thus, the questions raised for this investigation is as follows:

1. What is the nematode burden in flatfish in the inner and middle Oslofjord?
2. What are the preferred hosts for the nematode species at the investigated areas?
3. Does the nematode infestation have an impact on the condition of the fish?
4. What is the length and age distribution of the flatfish species in the inner and middle Oslofjord?

1.1 The biology of the nematodes

1.1.1 *Hysterothylacium aduncum* (Rudolphi, 1802)

The anisakid genus *Hysterothylacium* constitute over 50 species worldwide, however only one species of this genus is found in Norwegian waters, the *Hysterothylacium aduncum* (Berland 1961; Køie 1993; Anderson 2000). *H. aduncum* is found in a large number of marine teleost fish in temperate and cold waters (Berland 1989, 1998 & 1991; Andersen 1993). It is a widespread and abundant nematode parasite in the North Atlantic (Berland 1961; Andersen 1993) and it is believed that the reason for this is that *H. aduncum* mainly eats the stomach content and the hosts ingested prey, thus it is the food source that is of importance, not the host itself (Berland 1998). The nematodes mature in fish hosts and a third stage larva are encapsulated in the host's viscera. Though the life cycle is not fully known, there seems to be a general agreement that there are at least two intermediate hosts (Berland 1961; Andersen 1993; Køie 1993; Klimpel & Rückert 2005).

The life cycle of *H. aduncum* (Figure 1.1), begins when a young developing larva moults in the egg and a second stage larva develops of the surrounding cuticle from the first moult. A young third stage larva is developed by the shed of the cuticle from these two stages. Many various invertebrates act as the first (transport) host for the larvae, the egg might be taken up by small crustaceans, then hatch in their intestine and penetrate the hosts haemocoel. If this is a suitable host, the larvae will continue to grow, but since small crustaceans often are eaten up by larger animals such as other crustaceans, polychaetes, chaetognats, ctenophores, medusa and fish, these larvae will be carried up in the food chain (Køie 1993; Berland 1998; Anderson 2000). Maturation and reproduction take place in many fish species with different foraging patterns, but Gadoids are believed to be the final host (Berland 1989, 1998 & 1991; Andersen 1993). Thus the various fish species might act as transport hosts; this is when the third stage larvae are found in the viscera and the body cavity, or as the final host; when adult worms occur in the digestive tract. The life cycle is complete when the final host defecates and eggs from the parasite are shed in the ocean and can enter the cycle again if ingested by small crustaceans.

For the larvae to moult and become mature, certain physiological occurrences or sizes must be reached (Berland 1989). Køie (1993) found that certain length of the larvae decides the fate for further development. Larvae under 2 mm did not survive, whereas larvae between 2 – 3 mm did survive and could penetrate into the host's body cavity and be encapsulated there. When the larvae reaches over 3 mm in the first transport host (crustaceans or other invertebrates) and are ingested by a fish, the larvae will grow and moult twice and then migrate to the gut becoming an adult fifth stage larvae. Unlike most nematodes that can be found in restricted areas of the digestive tract, *H. aduncum* can be found throughout the entire gut. It can move freely in the stomach and intestine due to its alae (cervical wings). When the fish host dies, the larvae may move out of the host's mouth, gills and sometimes through the anus (Berland 1998; Anderson 2000).

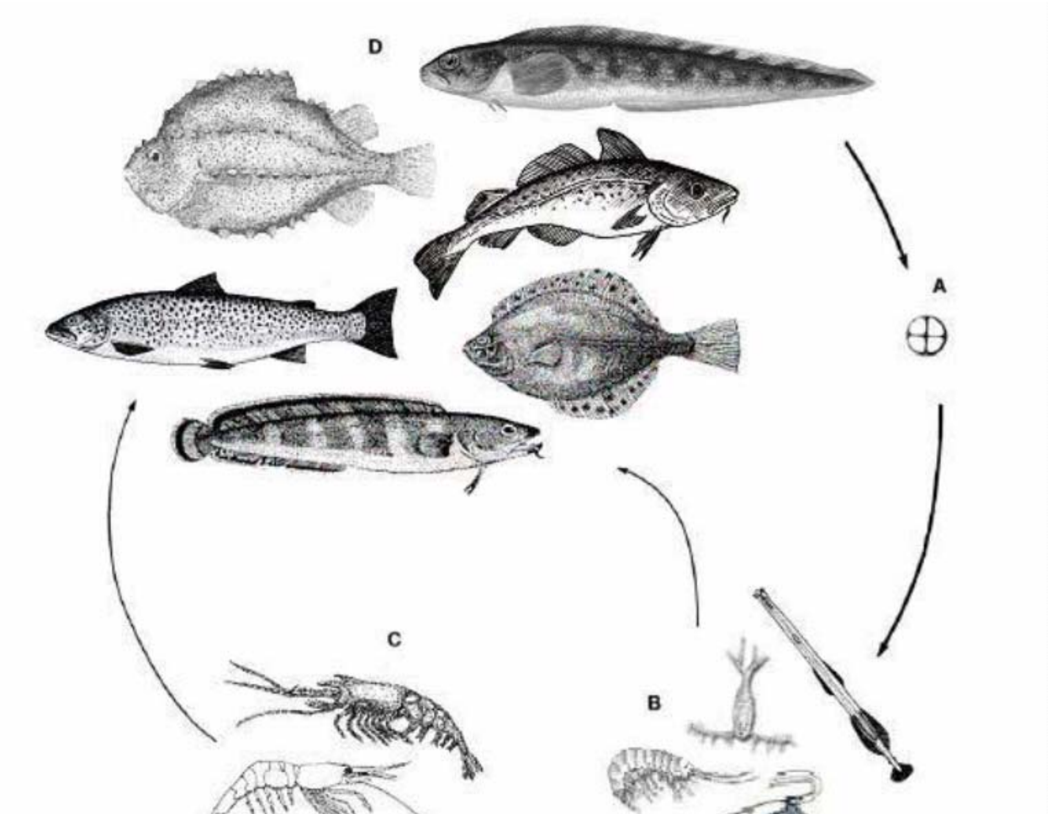

Figure 1.1. Life cycle of *Hysterothylacium aduncum*. A) Egg. B) First transport host (crustaceans and other zooplankton. C) Second transport host (larger crustaceans, zooplankton and fish). D) Final host (Fish). Reproduced from Hansen & Malmstrøm (2006).

1.1.2 *Cucullanus heterochrous* (Rudolphi, 1802)

Cucullanus heterochrous belongs to the family Cucullanidae and is mainly found in plaice and flounders (Pleuronectiformes) where they feed on the host's stomach content and the intestine (Berland 1961; Jensen 1991; Anderson 2000; Køie 2000). A recognized description of the development and transmission of cucullanids is still inadequate (Anderson 2000). The North-East Atlantic consists of two species of cucullanids: *C. cirratus* which generally infect gadoids and *C. heterochrous* which generally infect flatfish (Køie 2000a & 2000b).

The life cycle of *C. heterochrous* starts with eggs being discharged from a fish (Pleuronectiformes) into the water where they evolve to embryos (Figure 1.2). The embryos undergo three development stages; first (L1), second (L2) and third stage (L3), whereas the latter is the hatching stage. The embryo does not hatch until it is devoured by a transport/intermediate host. Polychaete specie, *Nereis diversicolor* seems to act as a true intermediate host in the North Atlantic (Køie 2000a). When the polychaete is infected, the larvae start to grow but do not develop any further. The cycle is complete when a suitable fish host (flatfish) eats the polychaete, the larvae may then develop to a fourth (L4) and fifth, the sexually mature, stage (L5). Thus, *C. heterochrous* found in flatfish are in their fourth or fifth larval stage.

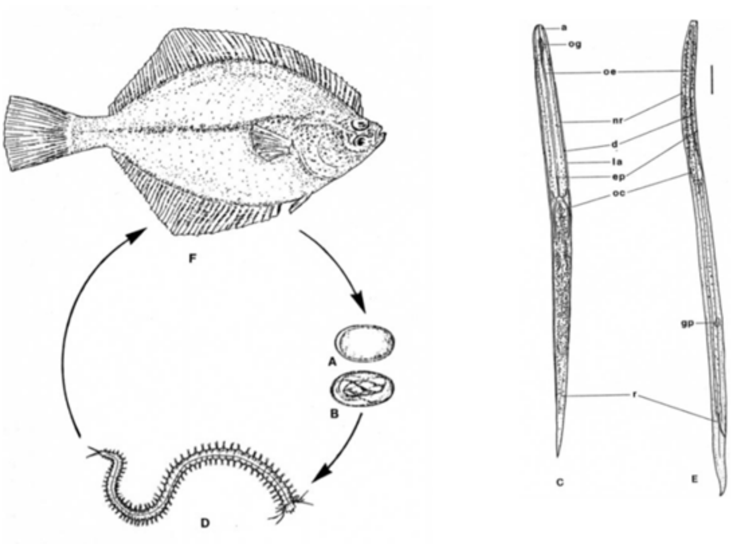

Figure 1.2. Life cycle of *Cucullanus heterochrous.* A) Fertilized newly shed egg. B) Embryonated, infective egg. C) Infective third-stage larvae (430 µm) from hatched egg. Ventral view. D) An intermediate host. E) Third-stage larvae (800µm) from intermediate host. Lateral view. F) Final host, flatfish (Køie 2000a).

1.1.3 Other nematodes

The three nematode species *Anisakis simples* (Rudolphi, 1809), *Pseudoterranova decipiens* (Krabbe, 1878) and *Contracaecum osculatum* (Rudolphi, 1802) mature in the stomach and intestine of marine mammals and birds in temperate and polar regions. Many intermediate hosts are used for the earlier life stages. *A. simplex* uses Euphausiids (krill) as a first intermediate host, while many teleost species are used as secondary hosts, before maturing in pinnipeds or cetaceans (Berland 1989). *P. decipiens* may use isopods, copepods or polychaetes as first intermediate host and various invertebrates and fish species might act as the second host before maturing in seals (McClelland 1990). *C. osculatum* has almost the same hosts and life cycles as *P. decipiens* (Anderson 2000).

1.1.4 Fish

The three species of flatfish focused on in this investigation was: American plaice (Fabricius, 1780), European plaice (Linnaeus, 1758) and Witch flounder (Linnaeus, 1758). These are all benthic species living on the soft bottom with a variable diet. Crustaceans, brittle stars, molluscs, some fish species and worms are the preferred food source. European plaice is found in depths less than 100 m and has the shallowest distribution of the three species. American plaice is found in depths between 10 and 400 m. Witch flounder is common in the deeper areas of the ocean floor, ranging down to 1460 m (Cargnelli *et* al. 1999; Hoarau *et* al. 2004).

2 Materials and methods

2.1 Study area

2.1.1. The Oslofjord

The Oslofjord (Figure 2.1) is 110km long and is located on the southeast part of Norway ranging from 59°01' N to 59°55' N and 10°15' E to 11°10' E (Andersen *et* al. 1970). It is separated into inner and outer Oslofjord by a bathymetric barrier, a sill at 19m depth near Drøbak. The Oslofjord is divided into five sections: (1) The outer Oslofjord from Færder to Horten/Moss, (2) the central basin, Breiangen, from north of Horten to Drøbak, (3) the Drøbak Sound, (4) the inner Oslofjord, including waters north of Håøya, the Vestfjord and the Bunnefjord and (5) the Drammensfjord which forms a separate system, connected to the western area of Breiangen (Webb *et* al. 2009). Three sections were investigated in my thesis project: Breiangen, the Drøbak Sound and the inner Oslofjord. Four stations (Midtmeie, Hellvik, Drøbak Sound and Tofte Plateau) were repeatedly sampled, while one station (Gråøya Through) was sampled once, all by trawling with the research vessel R/V Trygve Braarud in 2013 (Figure 2.1).

According to Gade (1963), the Skager Rack and the Oslofjord is separated by a ridge with a sill depth of about 120 meters. Inside this ridge the fjord extends to more than 360 meters depth. The outer Oslofjord is separated from Breiangen by yet another ridge of 110 meters; Breiangen itself is about 200 meter deep.

The island Håøya separates the inlet of the inner fjord in two. Bottom topography of the inner fjord is extremely variable with islands and skerries and minor ridges – these influence the deep water circulation of the fjord. The ridge between Bygdøy peninsula and Nesoddtangen is predominant in separating deep water in the Vestfjord from that in the Bunnefjord, sill depth here is about 50 meters.

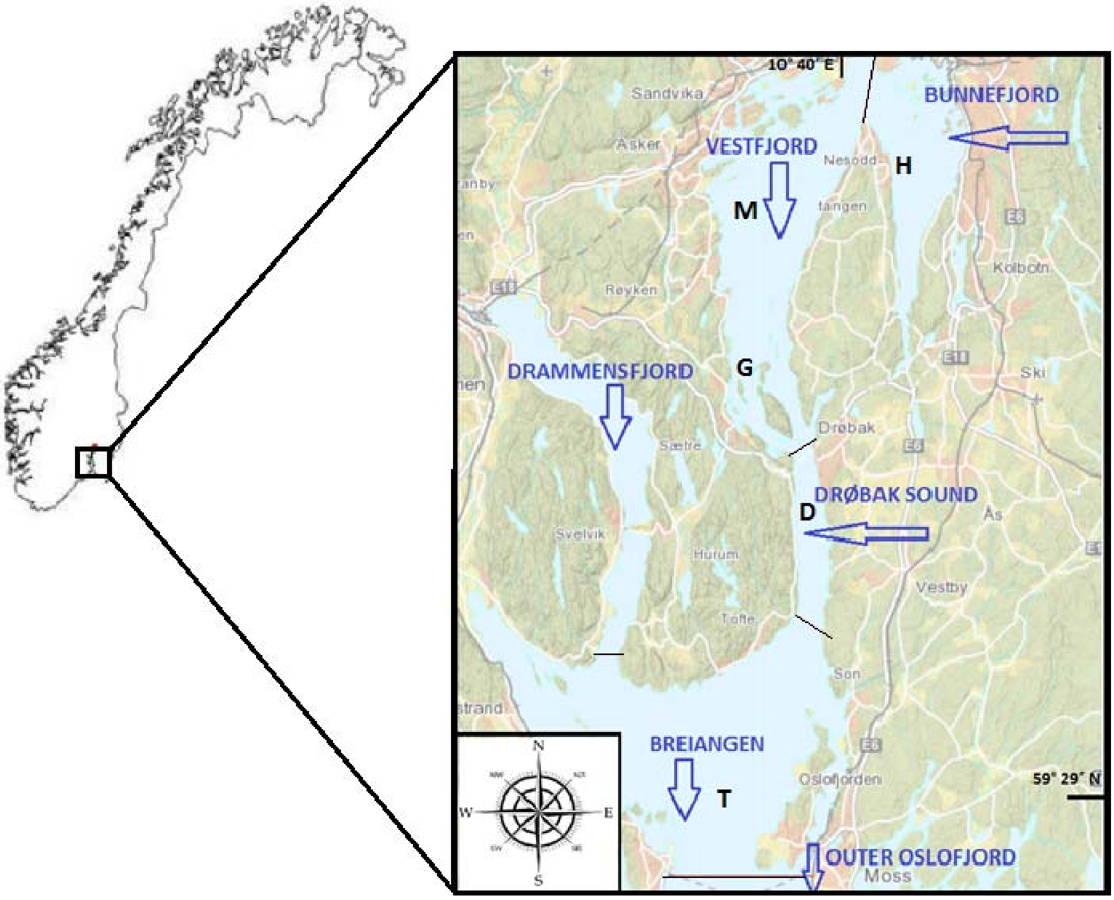

Figure 2.1. Map showing the five sections of the Oslofjord and the five sampling stations: Midtmeie (M), Hellvik (H), Drøbak Sound (D), Gråøya Through (G) and Tofte Plateau (T). Map made in www.norgeskart.no by Tonje Cecilie Urskog.

2.1.2. Midtmeie

Midtmeie is located southwest of the archipelago Steilene in the Inner Oslofjord. In total 12 trawling hauls at depth 80 to 100 meters were taken between March 2013 and November 2013. Sediment type is a mixture of mud and silt.

2.1.3. Hellvik

Hellvik is located on the eastern side of Nesoddtangen in the Inner Oslofjord. In total 4 trawling hauls at depth between 77 and 102 meters were taken between April 2013 and November 2013. Sediment type is a mixture of mud and silt.

2.1.4. Drøbak Sound

The Drøbak Sound is located by the town of Drøbak in the middle of Oslofjord and separates the Inner and the Outer Oslofjord by a sill. This sill restricts deep water circulation, exchange and renewal in the inner fjord (Gade 1963).

In total 3 trawling hauls at depth between 180 and 200 meters were taken between February 2013 and September 2013. Sediment type is a mixture of mud and silt.

2.1.5. Gråøya Through

Gråøya Through is located between Gråøy and Håøy in the inner Oslofjord. Only one trawling hauls at depth between 80 and 114 meters were taken in April 2013. Sediment type is a mixture of mud and silt.

2.1.6. Tofte Plateau

Tofte Plateau is located outside Horten and Moss in the Middle Oslofjord. In total 3 trawling hauls at depth between 120 and 140 meters were taken between October 2013 and December 2013. Sediment type is a mixture of mud and silt.

2.2 The fish samples

386 fish, belonging to 6 species, were sampled by trawling between 1 and 2 hours depending on the size of the fish steam and the bottom topography (usually between 70 to 110m). 383 fish was used in this project.

Due to time constrictions on the boat most fish were stored frozen in the laboratory at the University of Oslo before dissection. However, a few fish (8-10) were examined on board while the fish were still fresh and the nematodes alive. It is reasonable to believe that the worms are easier to see when alive during the dissection. According to my experience I believe that freezing the fish did not cause any sampling error in my analyses.

In the laboratory, the fish length was measured to the nearest centimeter and weight to the nearest gram, using a standard method of measurement. Fork length (FL) is one of the three measures commonly used on fish and was therefore the method for my thesis (Figure 2.2; Fisher *et* al. 1996; Kruse & Hubert 1997).

During fish dissection otoliths were removed for age determination and the body cavity, mouth and gastrointestinal were examined for nematodes.

Fish sex was determined by looking for gonads, mature fish tend to have very clear sexual difference, where females possess eggs and male possess sperm. Female tend to have more dark pink gonads, while males tended to have more white - to light pink gonads.

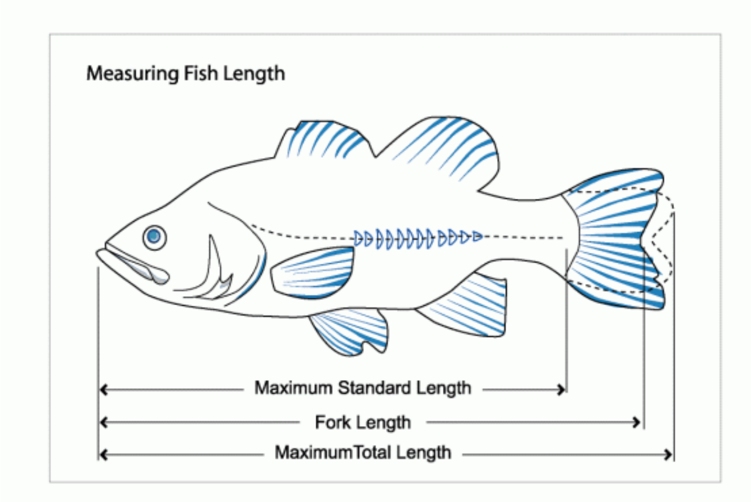

Figure 2.2. Measures of fish length. (http://www.fsl.orst.edu/geowater/FX3/help/9_Fish_Performance/Measures_of_Fish_Length.htm)

2.2.1. Age determination

The inner ear of teleost fish contains three pairs of otoliths composed of calcium carbonate: sagittal (which is the largest one) lapillus and asteriscus. Saggita is normally used for determining age because of its larger size, thereby used as a synonymous for otolith further on in this thesis.

The ear stones are located in a cavity filled with endolymph inside the fish skull (three cavities in total - Degens *et* al. 1969; Yoshinaga *et* al. 2000; Campana 2001). There are different polymorphs of calcium carbonate. Sagittae and lapilli are aragonite based, whilst asterisci are vaterite based, giving it its glassy appearance (Campana

1999). Sagittal otolith also includes otolin, a protein with high abundance of acidic amino acid.

According to Campana (1999) there are several physical factors that can change the calcification composition depending on the endolymphatic fluid surroundings:

- The pH in the endolymph can be changed; this means that the amount of bicarbonate ions inside the endolymph changes.
- There can be a reduced alkalinity inside the endolymph, this normally regulates the proton secretion through the saccular epithelium, and this may reduce the calcification rate.
- Temperature, also a factor that may influence the calcification rate.

These variations can change during seasons, which induce formation of two different annual growth rings or annulae. These two types are: a wide spread transparent zone (light zone in Figure 2.3), this is formed during times where food availability is good (August-November), and a narrow opaque zone (dark zone in Figure 2.3), formed during times where food availability is scarce (December-July) (Dannevig 1956; Morales-Nin 1992).

After the otoliths are cleaned in water and ethanol, the age of the fish can be determined by counting the annual zones under a light microscope (Figure 2.3; Nordeng & Jonsson 1978; Campana 1999; Yoshinaga *et* al. 2000). Some species of fish (i.e. *Gadus morhua, Merlangius merlangus, Pollachius virens)* have thicker otoliths that make the zones difficult to see. These must be burned by a Bohning alcohol burner for about two minutes and then cracked alongside the mid-line with a scalpel in water. This method was not needed in the present thesis.

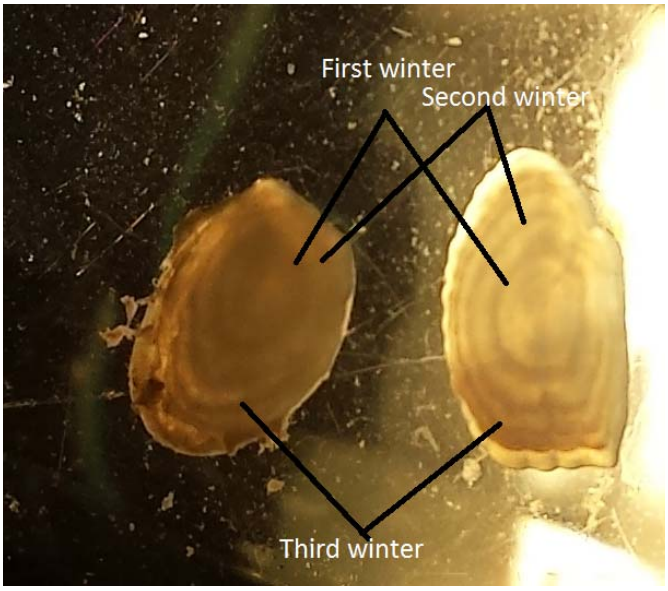

Figure 2.3. Growth zones on a sagittal otolith indicating age as number of years. Photo taken by Tonje Cecilie Urskog.

2.2.2. Nematode identification

I was taught to prepare and identify nematodes by Professor Emeritus Bjørn Berland from the University of Bergen and Karin Raamat, my co-supervisor.

Nematode species studied in the present thesis have five developmental stages. There are four larval stages (L1 to L4) and one adult (L5) stage, where only L3, L4 and L5 are found in fish. Identification of nematodes is complicated in the larval stages (L1 to L4) due to the lack of morphology that are not fully present yet (Perdiguero - Alonso *et* al. 2008).

Morphological characteristics for separating developmental stages are common among most nematode species (Aspholm 1991):

- L1 is larva enclosed in sheath of its host.
- L2 is larva molted in intestine, body cavity or intestine wall. Migration to mesenteries inside host.
- L3 is larva with boring tooth (depending on specie). Encysted larva, not enclosed in shed cuticle.
- L4 is larva with lips (depending on specie). Not possible to determine sex yet. Spicules, papillae or eggs not present.
- L5 is adult, female or male worms with lips. Spicules, papillae and eggs become visible. Reproductive organs are fully developed.

In the laboratory, the nematodes were fixed with Berland's fluid (consisting of 1 part of 40 % formalin and 19 parts of glacial acetic acid; Berland 1984) for a couple of minutes to uncoil and to become more transparent. This fluid also kills the worms quickly and increases them in length. The nematodes may be stored in Berland's fluid for a couple of days without disturbing or destroying the samples, but should preferably be stored in separate glasses with 70 % ethanol (Berland *pers. comm.*).

Nematode identification is primarily based on the structure of their internal organs (gastrointestinal system). Therefore it is necessary to make these organs visible under the microscope by using coloring liquids. Berland (1984) has shown that there are three different types of coloring liquids suitable for nematode identification, but as tested by Raamat (2012), lacto phenol (lactic acid, phenol, water and glycerol, ratio 1:1:1:1; with refractive index (RI) of 1:44) was sufficient for all nematodes in this project. According to Berland (1984, 2005) if penetrated by a suitable high RI, the tissue becomes transparent, helping identifying species.

In order to get the tissue more permeable for coloring liquids, the nematodes were put into acetic acid first. Bigger nematodes needed longer time in the acetic acid for better permeability, but about 3-10 minutes was usually enough. From the acetic acid nematodes were put on a microscopy glass with lacto phenol and cover slip on top of it.

After the identification, nematodes were placed back into the acetic acid to rinse them from the lacto phenol. This was necessary to avoid the nematodes from turning turgid (Berland 1961). Finally, the nematodes length was measured to the nearest millimetre and placed back to 70 % ethanol for storing. All nematodes were handled the same way for avoidance of any possible errors.

2.2.2.1 Identification of *Hysterothylacium aduncum*

Berland (1961, 1991, & 1998) and Anderson (2000) give a detailed description of the morphology and anatomy of *H. aduncum.*

Hysterothylacium aduncum has a very distinct characteristic tail, a "cactus tail" (Figure 2.4 a & d), present in L4 and L5 stages. The tail is conically shaped and both sexes have this. L3 larvae have a terminal bulb on its tail, and a cactus tail can be seen under the cuticle (Figure 2.4 c; Berland 1961, 1998). In L3, L4 and L5 stages the oesophagus (Figure 2.4 e & f) is long with a posterior appendix and an anterior intestinal caecum. The appendix and anterior caecum is of equal size. The excretory pore is at same level as the nerve ring (Figure 2.4 f), which is different from other species. Third stage larvae have a boring tooth ventrally placed to the mouth opening (Figure 2.4 f). Fourth and fifth stages have three lips with three semi-interlabia (Figure 2.4 e). Males are shorter than females, and the sexually mature males have spicules of equal size (Figure 2.4 a) and females have eggs (Figure 2.4 b). In L4 larvae you may see vulva, but papillae and spicules are not present yet (Berland 1961, 1998). It may be difficult to distinguish between L4 and a recently moulted L5.

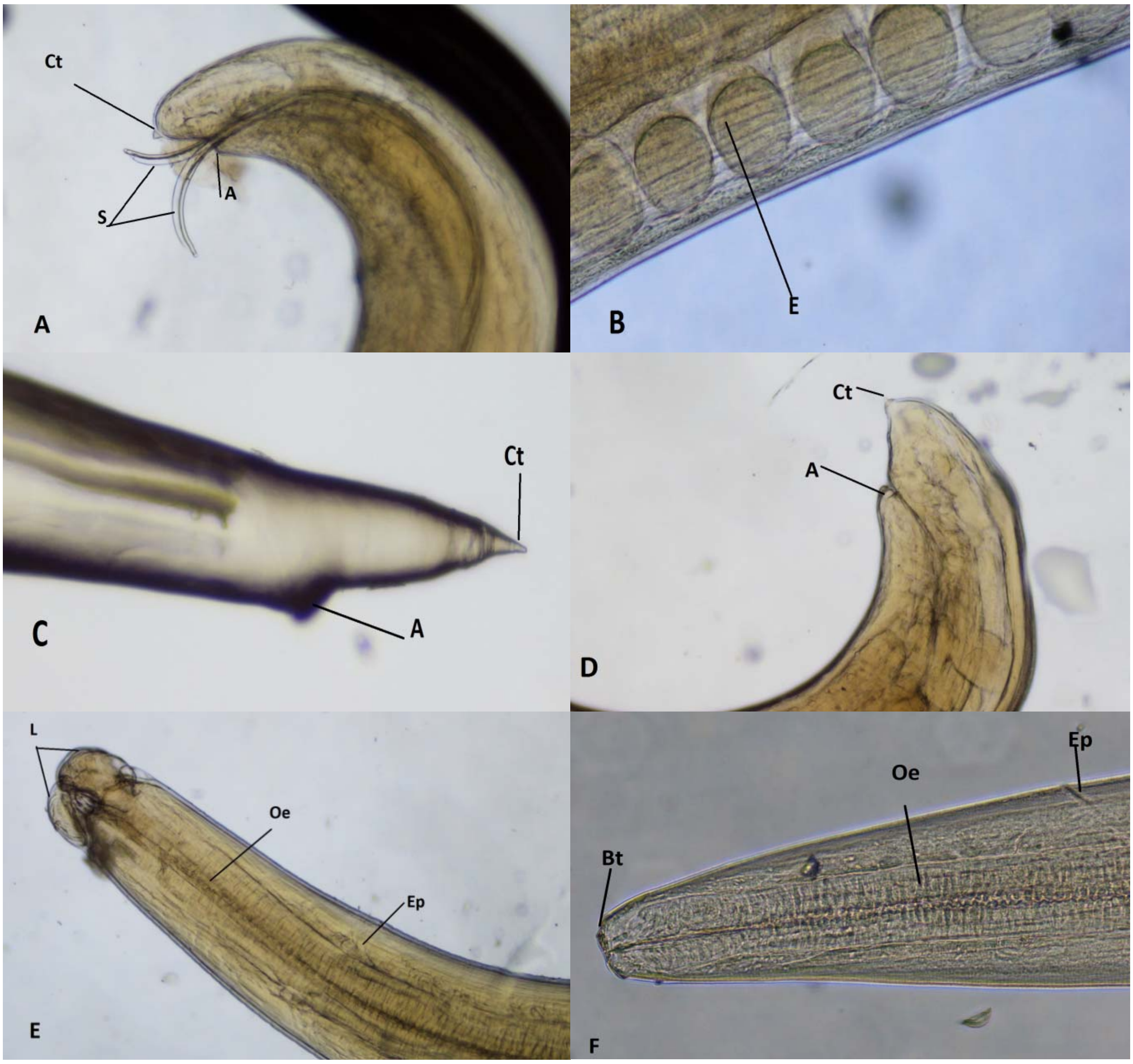

Figure 2.4. Different developmental stages of *Hysterothylacium aduncum.* **A** Posterior part of a fifth stage larva, male. Here seen with cactus tail (Ct), spicules (S) and anus (A), 4 x magnification. **B** Posterior part of a fifth stage larva, female with eggs (E) 4 x magnification. **C** Posterior part of a third stage larva with a cactus tail (Ct) seen under the cuticle and anus (A), 4 x magnification. **D** Posterior part of a fourth stage larva with cactus tail (Ct) and anus (A), 4 x magnification. **E** Head of a fourth stage larva, with two visible labia (L) with a visible semi-interlabia in between, oesophagus (Oe) and excretory pore (Ep), 4 x magnifications. **F** Head of a third stage larva, showing boring tooth (Bt), oesophagus (Oe) and excretory pore (Ep), 10 x magnification. Photo taken by Tonje Cecilie Urskog (2014).

2.2.2.2 Identification of *Cucullanus heterochrous*

A detailed description of the morphology and anatomy of *C. heterochrous* are made by Berland (1970) and Anderson (2000).

Cucullanus heterochrous is a medium-sized intestinal parasite normally found in plaice and flounders (Pleuronectiformes). It feeds on the stomach content and the abdominal wall. *C. heterochrous* has a characteristic head; it is bulb-shaped and quite straight (Figure 2.5 c). The parasite is white in color, hereby the name heterochrous, which differs from the color from another cucullanid, *cirratus,* which is more yellow (Berland 1970)*.* Oesophagus is short and a nerve ring (Figure 2.5 c) is situated about 1/3 to 1/2 of the parasites length from the anterior end. A simple intestine (Figure 2.5 d) with an excretory pore is placed a bit down the parasites body. The male is a bit shorter than the female. Female parasites have a conical shaped tail with a small pair of phasmid between anus and tail (Jensen 1991). In L5 stage it may be possible to see oval eggs (Figure 2.5 b) inside the body, which is a clear sex indicator. Male parasites have a more narrow tail with a pointed tip, with several papillae arranged around. They have a notable ventral sucker and long spicules (Figure 2.5 a). The spicules reaches in front of the ventral sucker and the spicules are used for reproductive purposes. The spicules are about equal in length, usually the right spicule is straighter, whilst the left is more coiled (Berland 1970; Anderson 2000).

Figure 2.5. Various developmental stages of *Cucullanus heterochrous.* **A** Posterior part of fifth stage larva, males with tail (T), spicules (S), Ventral sucker (Vs) and anus (A), 10 x magnifications. **B** Posterior part of a fifth stage larva, female with eggs (E), 10 x magnifications. **C** Head of a fifth stage larva, female with oesophagus (Oea) anterior side and posterior side (Oep) and nerve ring (Nr), 10 x magnifications. **D** Middle section of a fourth stage larva with intestinal caecum (InC) and intestine (Int), 10 x magnifications. Photo taken by Tonje Cecilie Urskog (2014).

2.3 Statistical methods

2.3.1 Characterization of infection rate

For describing the infestation rate of nematodes in fish, Margolis *et* al. (1982) suggested a standard terminology as following. Prevalence is defined as the fraction of infected fish, meaning the total number of fish containing at least one parasite divided by the total number of fish investigated. This is generally given in percentage of infected fish. Abundance is defined as the average number of parasites per investigated fish, and can be resolved by dividing the total number of parasites by the total number by the total number of investigated fish. Intensity is defined as the average number of parasites per investigated fish and can be resolved by dividing the total number of parasites by the total number of fish containing at least one parasite.

2.3.2 Condition factor

The condition factor, K = 100 W/L where W is the total body weight (g) and L is the total length (cm; Ricker 1975) is given for the health of fish by Williams (2000). Thus, this gives us a reason for investigating whether or not there is a relationship between the nematode burden and the condition of the fish. According to Froese (2006) median b shows the relationship between weight and length for a given specie in the equation of the condition factor, and if $b = 3$ small specimens in the samples have the same form and condition as large specimens. If $b > 3$ the larger specimens have increased in height or width more than length, and if $b < 3$ the larger specimens have changed their body shape, becoming more elongated or small specimens are in better condition. Froese (2006) also stated that the mean condition of specimens as well as the difference in condition between small and larger specimens varies between season, localities and year; this might result in difference in weight-length relationships.

3 Results

The fish samples in the Oslofjord contained 386 individuals from 6 different species, of which 61 individuals from 3 species were infected with nematode parasites. A complete list of all examined fish is given in Appendix I. There were some variations in the species compositions of fish at the five investigated stations. In the inner Oslofjord (Midtmeie, Hellvik and Gråøyrenna), American plaice represented 90.5 % (272 individuals), European plaice 4.5 % (13 individuals) and Witch flounder 5 % (15 individuals). In the Middle Oslofjord (Drøbak and Tofteflaket), American plaice represented 79.5 % (66 individuals), European plaice 12 % (10 individuals) and Witch flounder 8.5 % (7 individuals) (Figure 3.1; Appendix I).

Site comparison was only performed on American plaice (*Hippoglossoides platessoides*), European plaice (*Pleuronectes platessa*) and Witch flounder (*Glyptocephalus cynoglossus*), with especial focus on American plaice, since there were consistent large catch of only these three species at all five sampling areas (Figure 3.1). Only a few Lemon sole (*Microstomus kitt*), European flounder (*Platichthys flesus*) and Common dab (*Limanda limanda*) were found in all sampling areas, so these three species were not included in the analysis.

All statistical analysis is shown in Appendix V.

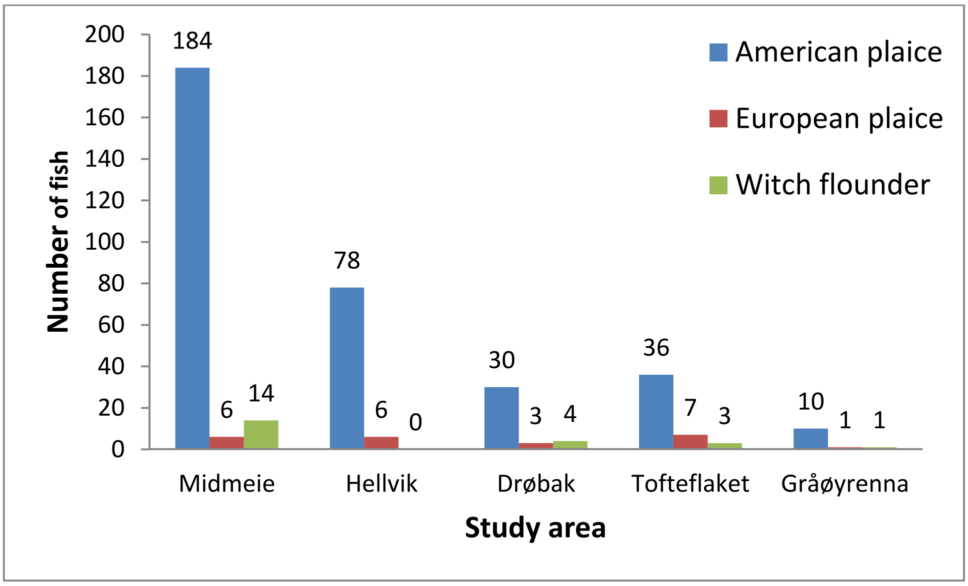

Figure 3.1. Number of American plaice, European plaice and Witch flounder caught at the five investigated stations.

3.1 Infestation rate

3.1.1 Prevalence

In the middle part of the Oslofjord (Tofteflaket and Drøbak) 44 out of 83 individuals (all three fish species) were infected by nematodes (all nematode species). In the inner part of the Oslofjord (Midtmeie, Hellvik and Gråøyrenna) only 17 out of 300 fish that were infected. *H. aduncum* had the highest prevalence of the two nematode species and were found on almost all locations except Gråøyrenna. In contrast, *C. heterochrous* was only found on two locations, Hellvik and Gråøyrenna.

American plaice had a fairly stable prevalence (16 %) in the Oslofjord, mostly with *H. aduncum*. European plaice was mainly infected by *C. heterochrous* and *H. aduncum* at Hellvik, Gråøyrenna and Tofteflaket. Witch flounder was only infected with *H. aduncum* at Drøbak and Tofteflaket.

In the middle part of the Oslofjord 44 fish out of 83 were infected by nematodes, while at the inner part of the Oslofjord only 17 fish out of 300 were infected. The highest prevalence was found at Tofteflaket: 74 % of all three species (American plaice, European plaice and Witch flounder) had at least one nematode inside. This location is situated in the Middle Oslofjord. Drøbak, which is located between the outer and the inner Oslofjord, had a prevalence of 27 % for all three species.

The three stations considered the Inner Oslofjord (Midtmeie, Hellvik and Gråøyrenna) had a prevalence of 5.4 %, 5.9 % and 8.3 % respectively. However, the sample sizes varied considerably between these three areas: 204, 84 and 12 respectively. See Appendix II for more detailed information.

In the total sample of all fish in this investigation, 14.9 % was infected with *H. aduncum* and 1 % with *C. heterochrous.* The prevalences of the nematode *H. aduncum* was 5.4 % (Midtmeie), 2.4 % (Hellvik), 27.0 % (Drøbak), 74 % (Tofteflaket) and 0 % (Gråøyrenna) and the prevalences for *C. heterochrous* were 0 % (Midtmeie), 3.6 % (Hellvik), 0 % (Drøbak), 0 % (Tofteflaket) and 8.3 % (Gråøyrenna).

Among the three species of flatfish there was just a little difference between the prevalences, 16 % in American plaice, 17 % in European plaice and 13.6 % in Witch flounder (Table 3.1a).

Compared to the other species, the catch of American plaice was almost 17 times larger (Figure 3.1). Among the 338 fish caught, 54 were infected with nematodes. In the Middle part of the Oslofjord, 39 of 66 American plaice were infected. In contrast, only 15 of 272 were infected in the inner part (see Appendix II for more details).

At Midtmeie 6 % of the American plaice were infected by *H. aduncum*, while none were infected by C*. heterochrous.* At Hellvik 2.5 % of the fish were infected both by *H. aduncum* and *C. heterochrous.* At Drøbak 30 % of the American plaice were infected by *H. aduncum* while none were infected by *C. heterochrous*. Tofteflaket had the highest prevalence of *H. aduncum*, 83 % of the fish from this location were infected by this nematode. None were infected by *C. heterochrous* in Tofteflaket (Table 3.1 b and c).

In total 23 European plaice were sampled from the Oslofjord. 10 fish from the Middle Oslofjord and 13 from the Inner Oslofjord. Overall, only 4 European plaice were infected with nematodes. *C. heterochrous* was found only in one fish at Hellvik and one fish at Gråøyrenna, prevalences 16.6 % and 100 %, respectively. *H. aduncum* was found only in two fish from Tofteflaket with 28.5 % prevalence. At Midtmeie and Drøbak no infected fish were found (Table 3.1 b and c).

In total 22 Witch flounders were sampled from the Oslofjord. 7 fish from the Middle Oslofjord and 15 from the Inner Oslofjord. Overall, only 3 Witch flounders were infected with nematodes. *H.aduncum* was found in one fish from Drøbak and two fish from Tofteflaket, prevalences 25 % and 66 %, respectively. *C. heterochrous* was not found from Witch flounder throughout the study area (Figure 3.1 b and c).

Table 3.1. Prevalences (% of infected fish) in American plaice, European plaice and Witch flounder at the five investigated stations. The prevalence of *Cucullanus heterochrous* and *Hysterothylacium aduncum* are given in separately in (b) and (c). The number of examined fish (n) is given in parenthesis.

a) Total prevalence	**American plaice**	**European plaice**	**Witch flounder**
Midtmeie	6 % (184)	0 % (6)	0 % (14)
Hellvik	5.1 % (78)	16.6 % (6)	0 % (0)
Drøbak	30 % (30)	0 % (3)	25 % (4)
Tofteflaket	83 % (36)	28.5 % (7)	66 % (3)
Gråøyrenna	0 % (10)	100 % (1)	0 % (1)
Total	16 % (338)	17.3 % (23)	13.6 % (22)

b) Prevalence *C. h.*	**American plaice**	**European plaice**	**Witch flounder**
Midtmeie	0 % (184)	0 % (6)	0 % (14)
Hellvik	2.5% (78)	16.6 % (6)	0 % (0)
Drøbak	0 % (30)	0 % (3)	0 % (4)
Tofteflaket	0 % (36)	0 % (7)	0 % (3)
Gråøyrenna	0 % (10)	100 % (1)	0 % (1)
Total	0.59 % (338)	8.7 % (23)	0 % (22)

c) Prevalence *H. a.*	**American plaice**	**European plaice**	**Witch flounder**
Midtmeie	6 % (184)	0 % (6)	0 % (14)
Hellvik	2.5 % (78)	0 % (6)	0 % (0)
Drøbak	30 % (30)	0 % (3)	25 % (4)
Tofteflaket	83 % (36)	28.5 % (7)	66 % (3)
Gråøyrenna	0 % (10)	0 % (1)	0 % (1)
Total	15 % (338)	8.7 % (23)	13.6 % (22)

3.1.2 Abundance

Among all three fish species in the middle part of the Oslofjord the number of nematodes per fish varied between 1.5 up to 2.6 (both nematode species included). In comparison, the abundance in the inner part of the Oslofjord

varied between 0.05 up to 0.9 nematodes per fish. *H. aduncum* had the highest abundance of the two nematode species and were found on all locations except Gråøyrenna. *C. heterochrous* was only found at Hellvik and Gråøyrenna. American plaice had abundance between 0 and 3.05 at the stations in the Oslofjord. Most of the fish were infected with *H. aduncum*, and only a few with *C. heterochrous*. Both nematode species were found in European plaice with a total abundance of 0.3 in Hellvik, 1.4 in Tofteflaket and one fish from Gråøyrenna with 11 worms. Witch flounder had only parasites at Drøbak and Tofteflaket, and were only infected by *H. aduncum*.

The abundance at Tofteflaket was the highest among all stations with 2.6 nematodes per fish (Appendix II). This was about 50 times higher than at Midtmeie which had the lowest abundance of all the stations (0.05). In comparison, Drøbak had an abundance of 1.6, Hellvik 0.1 and Gråøyrenna 0.9.

The abundance at Tofteflaket was the highest among all stations with 2.6 nematodes per fish (Appendix II). This was about 50 times higher than at Midtmeie which had the least abundance of all the stations (0.05). In comparison, Drøbak had an abundance of 1.6, Hellvik 0.1 and Gråøyrenna 0.9.

The nematode *H. aduncum* had the highest abundance on Tofteflaket (2.6) and Drøbak (1.6). At Midtmeie and Hellvik there were 0.05 and 0.02 nematodes per fish, respectively. At Gråøyrenna no *H. aduncum* were found. *C. heterochrous* had somewhat lower abundances 0.9 at Gråøyrenna and 0.1 at Hellvik. At the remaining stations (Midtmeie, Drøbak and Tofteflaket *C. heterochrous* was not found (See Appendix II for more information).

The abundance in American plaice at Tofteflaket was about 51 times larger than at Midtmeie (3.05 and 0.06 respectively; Table 3.2 a). At Drøbak the abundance was almost 2 nematodes per fish (1.9). At Hellvik the abundance rate was 0.11; neither of the two nematode species was observed at Gråøyrenna. At Tofteflaket and Drøbak, the abundances of *H. aduncum* were 3.05 and 1.9 respectively and no *C. heterochrous* was found at neither of the stations. The abundances of both parasite species were very low at Hellvik and Midtmeie (between 0 and 0.09).

European plaice was infected with parasites at three locations. At Tofteflaket only *H. aduncum* was observed to cause the infection with the abundance of 1.4. At Hellvik only *C. heterochrous* was found from the fish with the abundance of 0.3. At Gråøyrenna only one European plaice was sampled and this had 11 *C. heterochrous* worms in its digestive tract (Table 3.2 b and c).

Witch flounder was infected with parasites at two locations and only *H. aduncum* was found from these fish. At Drøbak the number of nematodes per fish was two times higher than at Tofteflaket (respectively 0.5 and 1; Table 3.2 b and c).

Table 3.2. Abundances (number of nematodes per examined fish) in American plaice, European plaice and Witch flounder at the five investigated stations. Abundances for *Cucullanus heterochrous* and *Hysterothylacium aduncum* are given separately in (b) and (c). The number of examined fish (n) is given in parenthesis.

a) Total abundance	American plaice	European plaice	Witch flounder
Midtmeie	0.06 (184)	0 (6)	0 (14)
Hellvik	0.11 (78)	0.33 (6)	0 (0)
Drøbak	1.9 (30)	0 (3)	0.5 (4)
Tofteflaket	3.05 (36)	1.43 (7)	1 (3)
Gråøyrenna	0 (10)	11 (1)	0 (1)
Total	0.55 (338)	1 (23)	0.28 (22)

b) Abundance *C. h.*	American plaice	European plaice	Witch flounder
Midtmeie	0 (184)	0 (6)	0 (14)
Hellvik	0.09 (78)	0.33 (6)	0 (0)
Drøbak	0 (30)	0 (3)	0 (4)
Tofteflaket	0 (36)	0 (7)	0 (3)
Gråøyrenna	0 (10)	11 (1)	0 (1)
Total	0.02 (338)	0.56 (23)	0 (22)

c) Abundance *H. a.*	American plaice	European plaice	Witch flounder
Midtmeie	0.06 (184)	0 (6)	0 (14)
Hellvik	0.025 (78)	0 (6)	0 (0)
Drøbak	1.9 (30)	0 (3)	0.5 (4)
Tofteflaket	3.05 (36)	1.43 (7)	1 (3)
Gråøyrenna	0 (10)	0 (1)	0 (1)
Total	0.53 (338)	0.44 (23)	0.28 (22)

3.1.3 Intensity

American plaice had about 6 times higher intensity at Drøbak than at Midtmeie. The nematode *H. aduncum* had the highest intensities at Drøbak and Tofteflaket (respectively 5.9 and 3.6), while *C. heterochrous* had the highest intensity at Hellvik (3.0).

Among all stations, the highest intensity was found at Gråøyrenna (11; three flatfish and two nematode species pooled), but this may be an artefact of a small sample size, since only one fish was infected in that location (Appendix II). In comparison, at Hellvik the number of nematodes per fish was 2.2, at Drøbak 5.9 and at Tofteflaket 3.6 (for all flatfish and nematode species).

All fish species summed, *H. aduncum* had the highest intensities at Drøbak and at Tofteflaket (respectively 5.9 and 3.6). Generally, the number of *C. heterochrous* per infected fish was low on all stations (See Appendix II for more information).

The intensity of both nematode species in American plaice at Drøbak was about 63 times larger than at Midtmeie (respectively 6.3 and 1; Table 3.3 a). At Tofteflaket and Hellvik the intensities were respectively 3.7 and 2.3 nematodes per infected fish.

In European plaice and Witch flounder only *H. aduncum* was found in Tofteflaket, with intensities of respectively 5 and 1.5. At Drøbak the intensity were 2 in Witch flounder (Table 3.3 c). For *C. heterochrous* in European plaice at Hellvik and Gråøyrenna the intensity were 2 and 11 respectively (Table 3.3 b).

Table 3.3. Intensities (number of all nematodes per examined fish) in American plaice, European plaice and Witch flounder at the five investigated stations. The intensities of *Cucullanus heterochrous* and *Hysterothylacium aduncum* are given separately in (b) and (c). The number of examined fish (n) is given in parenthesis.

a) Total intensity	**American plaice**	**European plaice**	**Witch flounder**
Midtmeie	1 (11)	0 (0)	0 (0)
Hellvik	2.25 (4)	2 (1)	0 (0)
Drøbak	6.33 (9)	0 (0)	2 (1)
Tofteflaket	3.66 (30)	5 (2)	1.5 (2)
Gråøyrenna	0 (0)	11 (1)	0 (0)
Total	3.46 (54)	5.75 (4)	1.66 (3)

b) Intensity *C. h.*	**American plaice**	**European plaice**	**Witch flounder**
Midtmeie	0 (0)	0 (0)	0 (0)
Hellvik	3.5 (2)	2 (1)	0 (0)
Drøbak	0 (0)	0 (0)	0 (0)
Tofteflaket	0 (0)	0 (0)	0 (0)
Gråøyrenna	0 (0)	11 (1)	0 (0)
Total	3.5 (2)	6.5 (2)	0 (0)

c) Intensity *H. a.*	**American plaice**	**European plaice**	**Witch flounder**
Midtmeie	1 (11)	0 (0)	0 (0)
Hellvik	1 (2)	0 (0)	0 (0)
Drøbak	6.33 (9)	0 (0)	2 (1)
Tofteflaket	3.66 (30)	5 (2)	1.5 (2)
Gråøyrenna	0 (0)	0 (0)	0 (0)
Total	3.46 (52)	5 (2)	1.66 (3)

3.2 Infestation of *Hysterothylacium aduncum*

3.2.1 Infection in all samples of American plaice

Figure 3.2 shows the number of American plaice (most abundant fish specie) infected with *H. aduncum* for each year class and both genders throughout all study areas. The most infected year classes in both sexes were 2 and 3, with the abundances of respectively 13 and 10 in females and 8 and 4 in males. More detailed statistics for both sexes are given in Appendix IV.

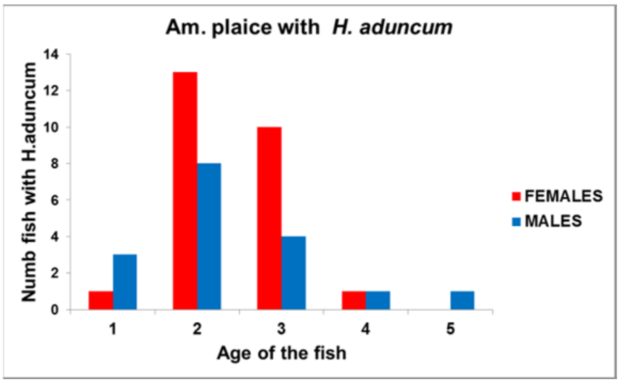

Figure 3.2. Number of American plaice infected with *Hysterothylacium aduncum* at each year class in females and males at all five investigated stations.

Figure 3.3 shows the prevalence of *H. aduncum* in female and male American plaice at 4 stations (Drøbak, Midtmeie, Hellvik and Tofteflaket). At Gråøyrenna no *H. aduncum* was found. Tofteflaket had clearly the largest percentage of infected fish, 85 % of females and 71 % of males were infected. Drøbak had the next largest prevalence of infected fish, with 35 % of the females and 14 % of the males infected. These locations are both located in the Middle of the Oslofjord. A figure of the abundance of *H. aduncum* in American plaice is given in Appendix IV.

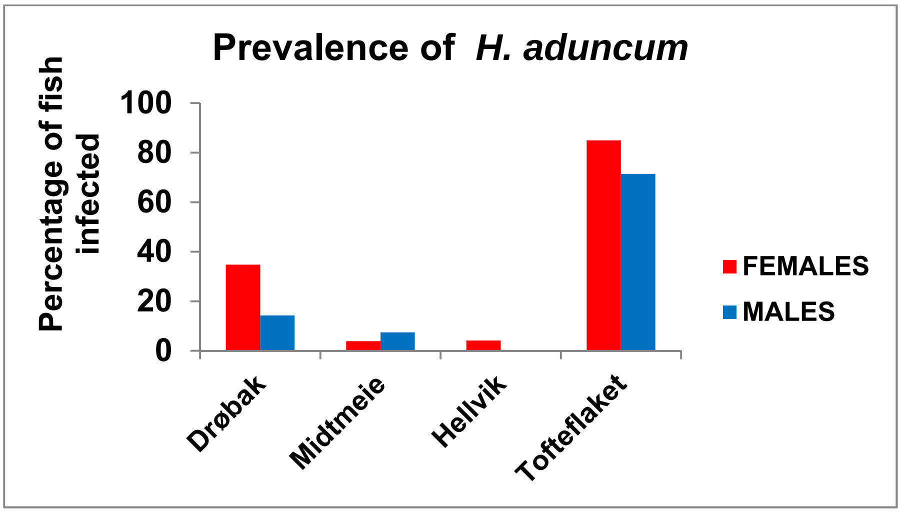

Figure 3.3. Prevalence of *Hysterothylacium aduncum* in females and males of American plaice at 4 stations (no H. aduncum was found at Gråøyrenna).

3.2.2 Infection in American plaice at the different locations

At Midtmeie 184 American plaice were caught; 77 females, 94 males and 13 juveniles (Appendix II). Of these 3.9 % of the females and 7.4 % of males were infected by *H. aduncum*, with the abundances of respectively 0.04 and 0.07 in females and males (all statistics are given in Table 3.4).

Table 3.4. Summary of the *Hysterothylacium aduncum* infection in American plaice at Midtmeie.

MIDTMEIE	FEMALES	MALES
Numb. Fish	77	94
Numb. inf. Fish	3	7
Numb. *H. a.*	3	7
Prevalence	3.9	7.4
Abundance	0.04	0.07
Intensity	1.0	1.0

At Hellvik 78 American plaice were caught; 24 females, 43 males and 11 juveniles (Appendix II). Of these only one fish, a female, was infected by *H. aduncum* and the corresponding prevalence was 4.2. No males were infected (all statistics are given in Table 3.5)

Table 3.5. Summary of the *Hysterothylacium aduncum* infection in American plaice at Hellvik.

HELLVIK	FEMALES	MALES
Numb. Fish	24	43
Numb. inf. fish	1	0
Numb. *H. a.*	1	0
Prevalence	4.2	0.0
Abundance	0.04	0.00
Intensity	1.0	0.00

At Drøbak 30 American plaice were caught; 23 females and 7 males (Appendix II). Of these, 34 % of the females and 14.3 % of the males were infected by *H. aduncum*, with the abundances of respectively 2.4 and 0.4 in females and males (all statistics are given in Table 3.6).

Table 3.6. Summary of the *Hysterothylacium aduncum* infection in American plaice at Drøbak.

DRØBAK	FEMALES	MALES
Numb. Fish	23	7
Numb. inf. Fish	8	1
Numb. *H. a.*	54	3
Prevalence	34.8	14.3
Abundance	2.35	0.43
Intensity	6.8	3.0

At Tofteflaket 36 American plaice were caught; 20 females, 14 males and 2 juveniles (Appendix II). Of these, 85 % of the females and 71 % of males were infected by *H. aduncum*, with the abundances of respectively 3.4 and 1.9 in females and males (all statistics are given in Table 3.7).

Table 3.7. Summary of the *Hysterothylacium aduncum* infection in American plaice at Tofteflaket.

TOFTEFLAKET	FEMALES	MALES
Numb. Fish	20	14
Numb. inf. Fish	17	10
Numb. *H. a.*	68	26
Prevalence	85.0	71.4
Abundance	3.40	1.86
Intensity	4.0	2.6

At Gråøyrenna 10 American plaice were caught; one female and 9 male (Appendix I). Of these, no fish were infected by *H. aduncum*.

3.3 Infestation of *Cucullanus heterochrous*

3.3.1 Infection in all samples of American plaice

Figure 3.4 shows the number of individuals infected by the nematode *C. heterochrous* at each age in female and male American plaice at all five stations. Only one American plaice were infected, a 2 year old male. Thus, the abundance for *C. heterochrous* was 0.16 at Hellvik. More detailed statistics for both sexes are given in Appendix IV.

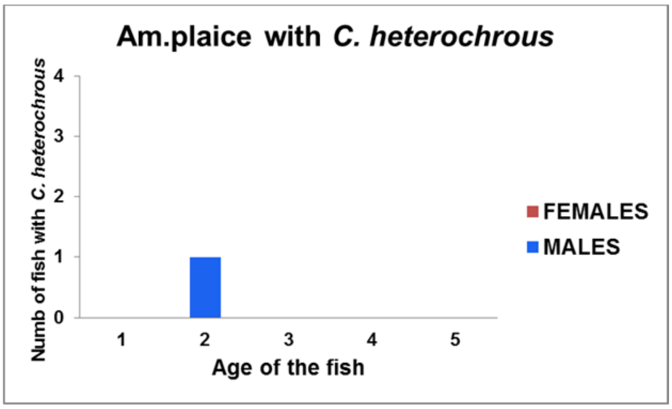

Figure 3.4. The distribution of the number of *Cucullanus heterochrous* at each age in female and male American plaice in all samples at all five stations.

Figure 3.5 shows the prevalence of *C. heterochrous* in female and male American plaice at all five stations. This nematode was only found at Hellvik where the prevalence was 0.04.

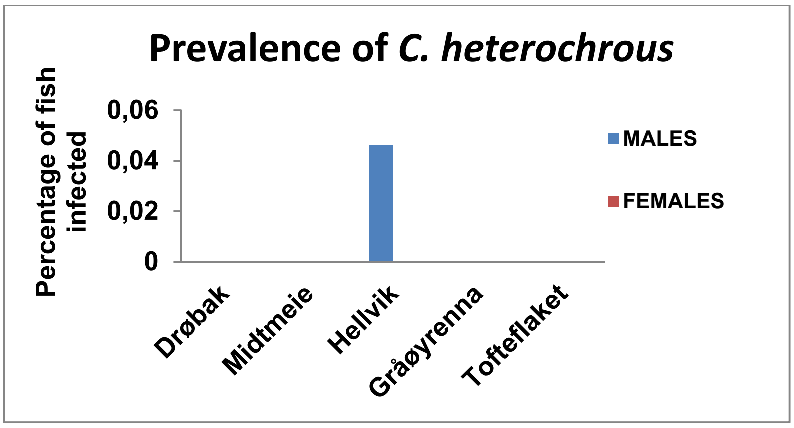

Figure 3.5. Prevalence of *Cucullanus heterochrous* in females and males of American plaice at all five stations. There was only infected fish in the total sample, and this individual was caught in Hellvik.

3.3.2 Infection in all samples of European plaice

Figure 3.6 shows the number of individuals infected by the nematode *C. heterochrous* at each age in female and male European plaice at all five stations. Only two European plaice were infected; two males at the age 3. One was caught in Hellvik, the other in Gråøyrenna giving abundances of respectively 0.66 and 11. More detailed statistics for both sexes are given in Appendix IV.

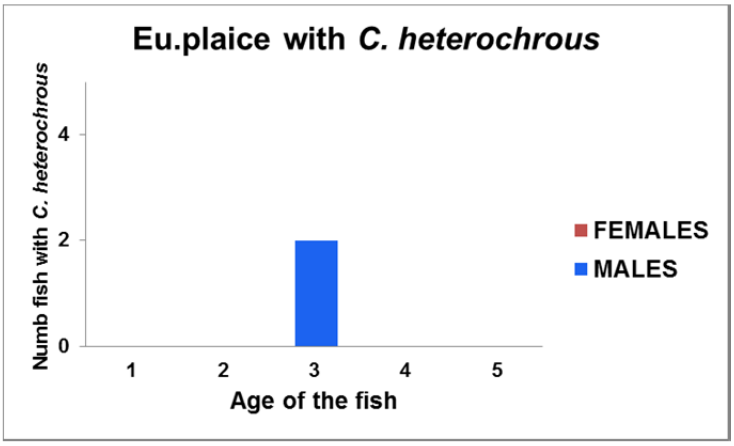

Figure 3.6. The distribution of the number of *Cucullanus heterochrous* at each age in female and male American plaice in all samples at all five stations.

Figure 3.7 shows the prevalence of *C. heterochrous* in female and male European plaice at all five stations. This nematode was only found at Hellvik and Gråøyrenna where the prevalence was respectively 0.33 and 1.

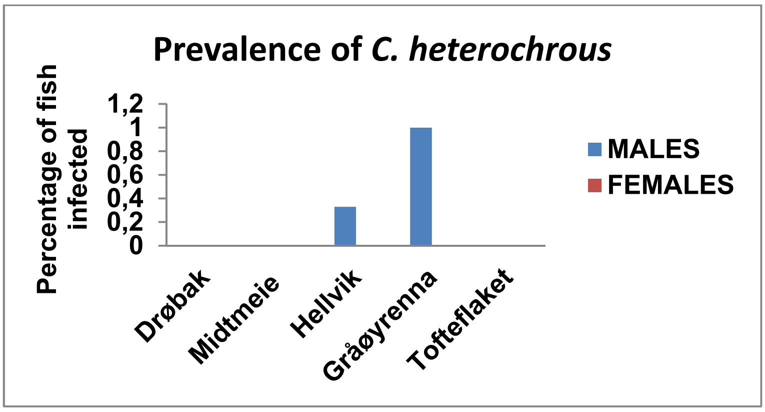

Figure 3.7. Prevalence of *Cucullanus heterochrous* in females and males of European plaice at all five stations. Infected fish were only found at Hellvik and Gråøyrenna.

3.3.3 Parameters in American plaice

At Hellvik 78 American plaice were caught; 24 female, 44 male and 10 juveniles (Appendix II). Of these, two fish were infected by *C. heterochrous*, a male (5 parasites) and one juvenile with 2 nematodes (Table 3.8).

Table 3.8. Summary of the *Cucullanus heterochrous* infection in American plaice at Hellvik.

HELLVIK	FEMALES	MALES
Numb. fish	24	44
Numb. inf. fish	0	1
Numb. *C. h.*	0	5
Prevalence	0	0.04
Abundance	0	0.11
Intensity	0	5

3.3.4 Parameters in European plaice

At Hellvik 6 European plaice were caught; 3 females and 3 males (Appendix II). Of these, only one male were infected by *C. heterochrous* (2 parasites - Table 3.9).

Table 3.9. Summary of the *Cucullanus heterochrous* infection in European plaice at Hellvik.

HELLVIK	FEMALES	MALES
Numb. Fish	3	3
Numb. inf. Fish	0	1
Numb. *C. h.*	0	2
Prevalence	0	3
Abundance	0	0.66
Intensity	0	2

At Gråøyrenna only one European plaice were caught; a male with 11 parasites (Appendix II; Table 3.10).

Table 3.10. Summary of the *Cucullanus heterochrous* infection in European plaice at Gråøyrenna.

GRÅØYRENNA	FEMALES	MALES
Numb. Fish	0	1
Numb. inf. Fish	0	1
Numb. *C. h.*	0	11
Prevalence	0	1
Abundance	0	11
Intensity	0	11

3.4 Nematode length

3.4.1 Average length for *Hysterothylacium aduncum*

Hysterothylacium aduncum were found at all stations except at Gråøyrenna, Table 3.11 shows the average length (mm) for this nematode at the four stations. The average length for all stations was 39.7 mm, where Tofteflaket had the highest average length of 43.2 mm. Hellvik had the shortest average length (20 mm), however, only two nematodes were found here.

Table 3.11. Average length of *Hysterothylacium aduncum* at Midtmeie, Hellvik, Drøbak and Tofteflaket.

Area	Average length (mm)
Midtmeie	29.2
Hellvik	20
Drøbak	34.8
Tofteflaket	43.2
Total for all areas	39.7

Table 3.12 shows the average length (mm) of *H. aduncum* in American plaice, European plaice and Witch flounder at the four stations. European plaice had the highest average length of 69.8 mm, while American plaice had an average of 37.6 mm.

Table 3.12. Average length of *Hysterothylacium aduncum* in American plaice, European plaice and Witch flounder

Fish specie	Average length (mm)
American plaice	37.6
European plaice	69.8
Witch flounder	51.6

3.4.2 Average length for *Cucullanus heterochrous*

Cucullanus heterochrous were found at Gråøyrenna and Hellvik, Table 3.13 shows the average length (mm) for this nematode at the two stations. The average length for all stations was 10.3 mm, where Gråøyrenna had the highest average length of 10.9 mm, while Hellvik had an average of 9.5 mm.

Table 3.13. Average length of *Cucullanus heterochrous* at Gråøyrenna and Hellvik.

Area	Average length (mm)
Gråøyrenna	10.9
Hellvik	9.5
Total for all areas	10.3

Table 3.14 shows the average length (mm) of *C. heterochrous* in American plaice and European plaice at the two stations. European plaice had the highest average length of 11.2 mm, while American plaice had an average of 8.5 mm.

Table 3.14. Average length of *Cucullanus heterochrous* in American plaice and European plaice.

Fish specie	Average length (mm)
American plaice	8.5
European plaice	11.2

3.5 Nematode length as a factor

Figure 3.8 shows the length of *H. aduncum* versus length (right column) and age (left column) of American plaice at all stations. It is seen that the year classes 2 and 3 were mostly infected by *H. aduncum* in both sexes and that there is no relationship between length of *H. aduncum* and the length and age of the host.

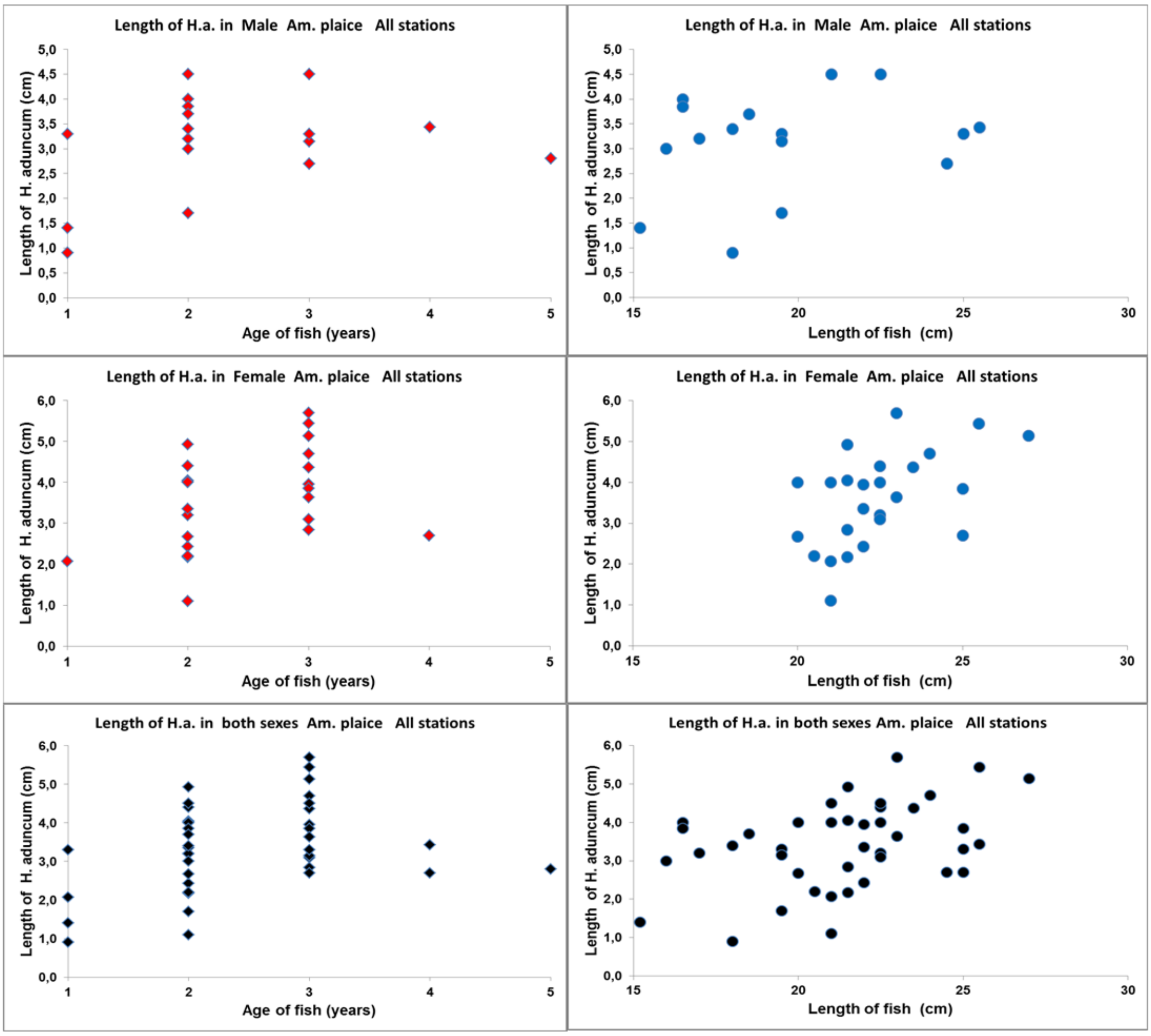

Figure 3.8. Length of Hysterothylacium aduncum vs the length and age of American plaice at all five investigated stations.

Figure 3.9 shows the length of *H. aduncum* versus length (right column) and age (left column) of American plaice at Tofteflaket which is the station with the highest abundance of this nematode. It is seen that the year classes 2 and 3 are heavily infected by *H. aduncum* in both sexes and that there is no relationship between the length of *H. aduncum* and the length and age of the host.

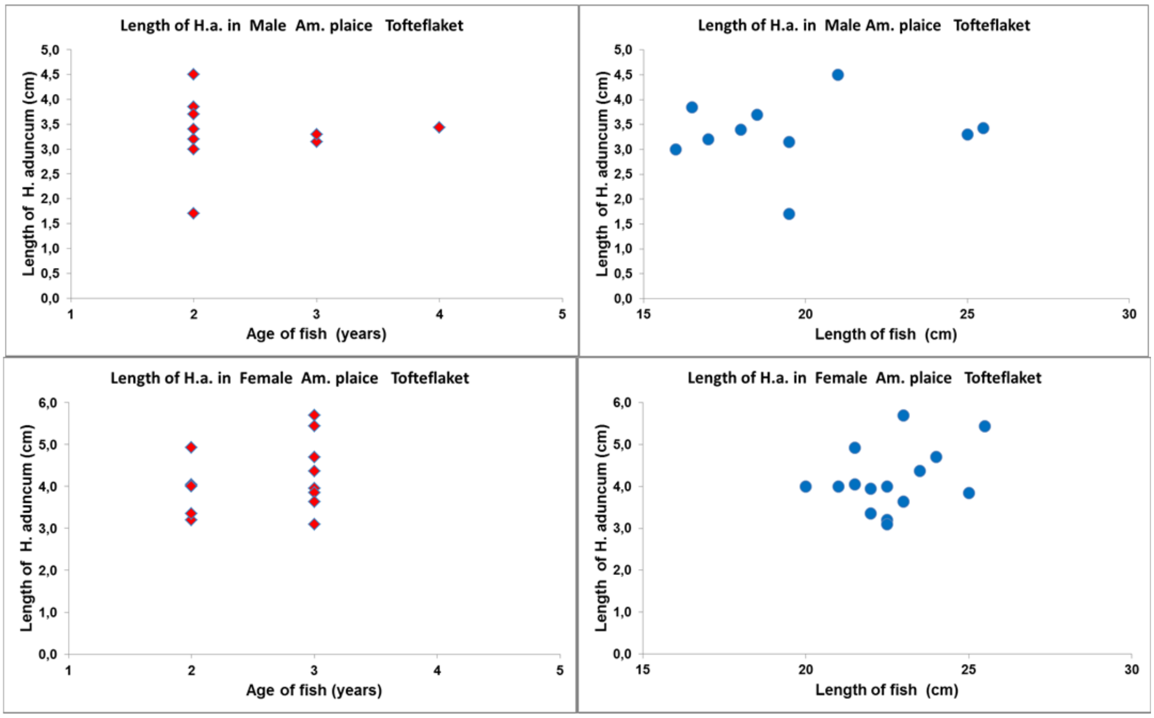

Figure 3.9. Length of *Hysterothylacium aduncum* vs age and length in female and male American plaice at Tofteflaket, the station with the highest abundance of *H. aduncum*.

3.6 Biological information of fish

The most predominant year class of the fish were 1- 3, and there was a large covariance between the length and age of the fish.

3.6.1 Age distribution in American plaice

Midtmeie

The most abundant fish in the samples was American plaice (338 individuals), and most of them was caught at Midtmeie (184 fish – Appendix II); where the year classes 1-3 dominated, and the sex ration was 77:94 females to males (13 juveniles were not included here).

There were few fish caught in the age groups 4 and 5 for both genders (a total of 14 %; Figure 3.10), of these only 3 % were males. Among 2 years old fish, 44 % were females and 31 % males (Figure 3.10).

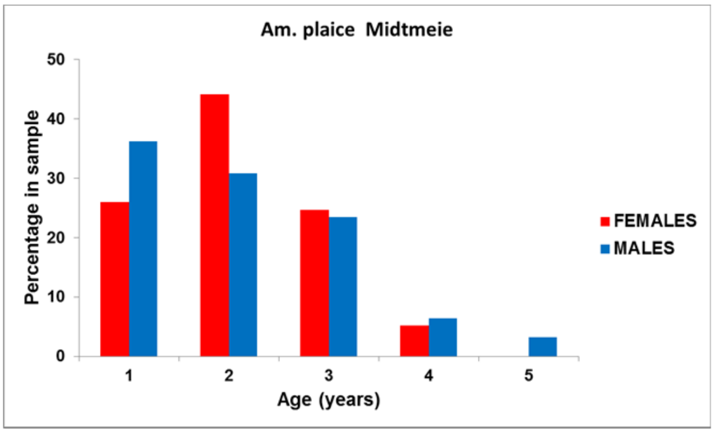

Figure 3.10. The age distribution (percentage) of female and male American plaice at Midtmeie.

Hellvik

At Hellvik, 78 American plaice was caught (Appendix II). Of these, the year classes were predominantly 1 to 3, and more males (43 individuals) than females (24 individuals) were caught (11 juveniles were not included here).

At 2 years, the percentage of males and females was almost the same (46 % of the females were found in this year class). For males, year class 1 and 2 were almost represented by the same percentage, 42 % and 47 % respectively. Also among females were the year classes 1 and 3 almost represented by the same percentage, 21 % and 25 % respectively (Figure 3.11).

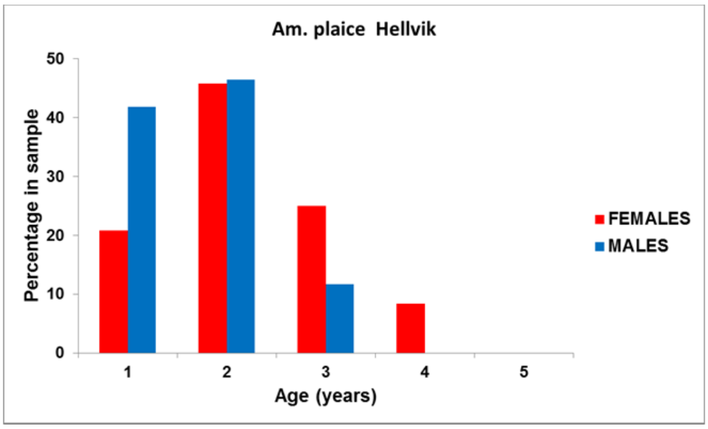

Figure 3.11. The age distribution (percentage) of female and male American plaice at Hellvik.

Drøbak

At Drøbak 30 American plaice were caught (Appendix II). Of these, there were more females (23 individuals) than males (7 individuals). No individuals were 4 or 5 years old. The percentage of females at 2 and 3 years were 52 % and 39 % respectively. The percentage for males at 1 and 2 years were 29 % and 71 % respectively (Figure 3.12).

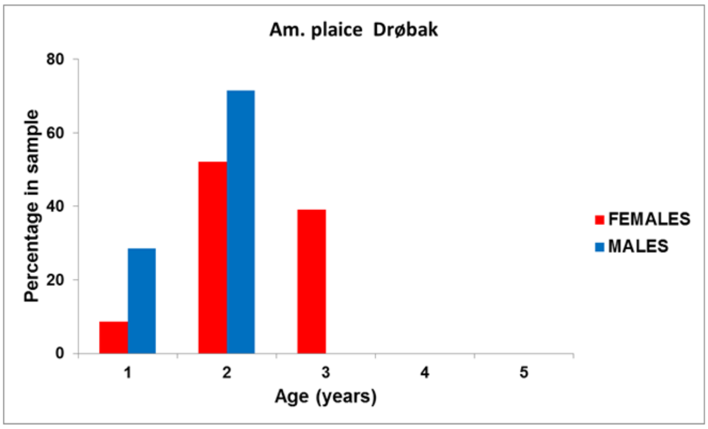

Figure 3.12. The age distribution (percentage) of female and male American plaice at Drøbak.

Tofteflaket

At Tofteflaket 36 American plaice were caught; 20 individuals were females and 14 were males (2 juveniles were not included here – Appendix II). None of the individuals of both sexes belonged to the age groups 1 and 5 (Figure 3.13). The percentage of females at 2 and 3 years were 40 and 50 % respectively. For males the percentage for 2 and 3 years were 71 % and 21 % respectively.

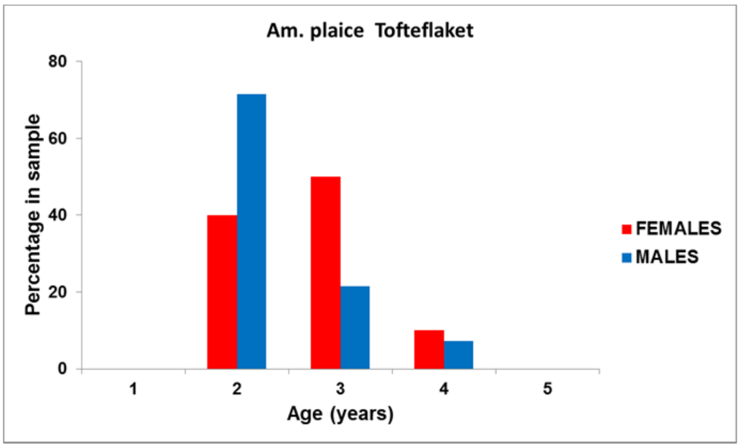

Figure 3.13. The age distribution (percentage) of female and male American plaice at Tofteflaket.

Gråøyrenna

At Gråøyrenna 10 American plaice were caught, of which 9 individuals were male (only one juvenile, this was not included here – Appendix II), and 44 % of them were 2 years (Figure 3.14).

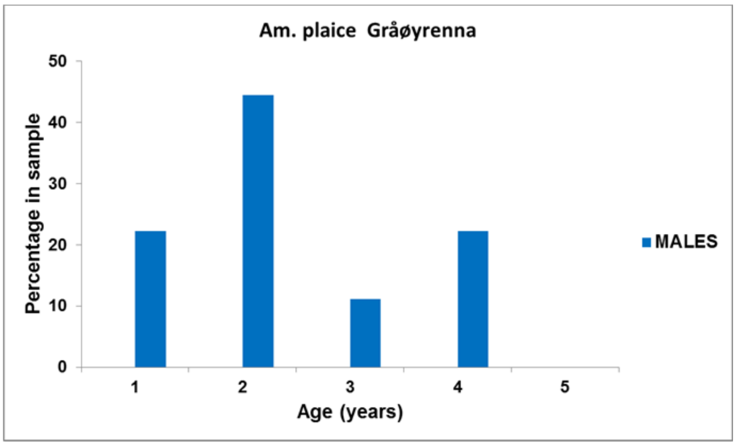

Figure 3.14. The age distribution (percentage) of female and male American plaice at Gråøyrenna.

3.6.2 Length vs age

Figure 3.15 shows the length of the fish (cm) at each age for American plaice. R^2 varied between 27 % and 70 % for American plaice at the five investigated stations. For females at Midtmeie, only 46 % of the variability in length was explained by age. For males this was 64 %. At Hellvik age could explain 66% of the variability in length for females and 55 % in males. At Drøbak corresponding numbers were 40 % females and 31 % for males.

For males at Tofteflaket, only 27 % of the variability in length was explained by age. For females this was 58 %. At Gråøyrenna age could explain 69 % of the variability in length for males.

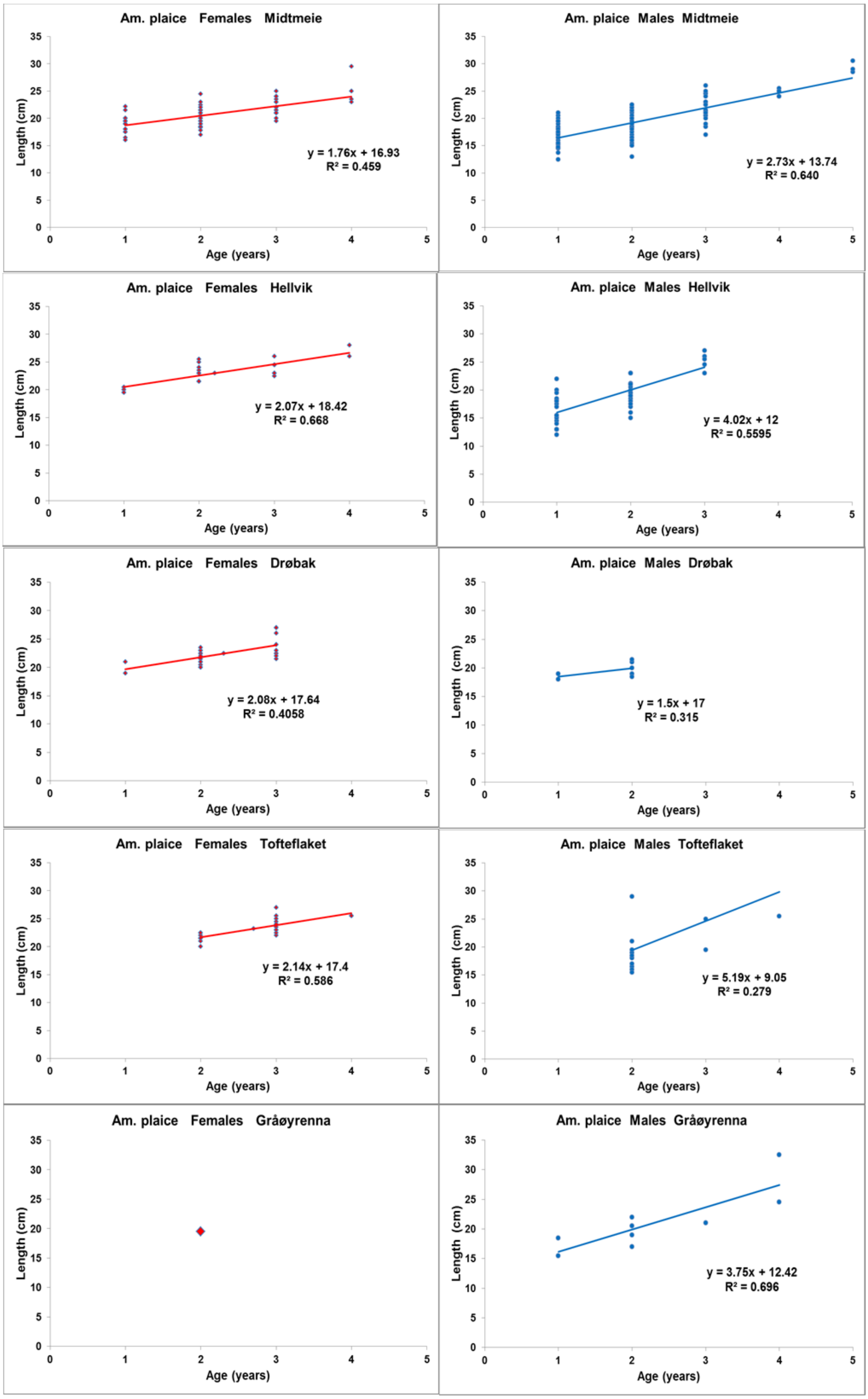

Figure 3.15. Length versus age for female and male American plaice at five stations.

3.6.3 Condition factor

Figure 3.16 shows the logarithmic relationship between the length (cm) and weight (g) in American plaice. R^2 varied between 75 % and 99 %. Thus, there is a high covariability between weight and length for female American plaice at all five stations.

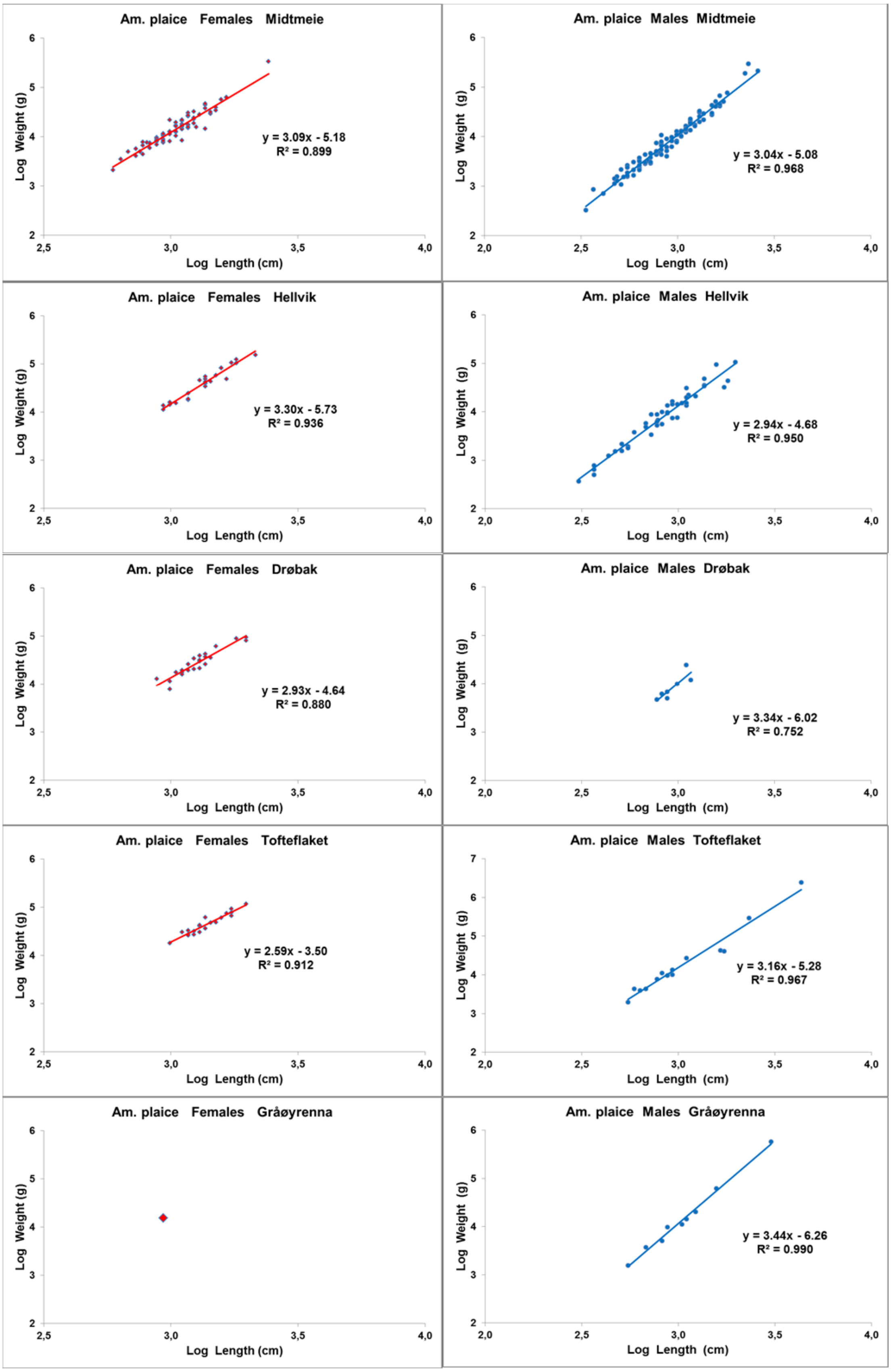

Figure 3.16. Weight vs length (on a Logarithmic scale) in female and male American plaice at five stations.

4 Discussion

During this project in the Oslofjord 386 fish belonging to 6 species were sampled by trawling at depths between 70 and 110 m at 5 locations (Midtmeie, Hellvik, Drøbak, Tofteflaket and Gråøyrenna). American plaice, European plaice and Witch flounder were caught at consistent numbers at all stations. Three other species, Lemon sole, European flounder and Common dab, only occurred in very low numbers (one of each) in the samples. American plaice was clearly the most abundant species in all samples.

Previous studies have mostly conducted sampling in the outer Oslofjord, where the nematodes *A. simplex* and *P.* decipiens which reproduce in marine mammals, occur with large abundances. In my investigation *H. aduncum* was the most abundant nematode in flatfish in both the inner and the middle Oslofjord. *H. aduncum* has been an increasing parasite over the years, and Raamat (2012) found it to be the most widespread nematode in her investigation at Torbjørnskjær and Langøya. Hansen & Malmstrøm (2006) stated that the abundance of this nematode was decreasing and explained this decline by a change of host because cod-fish decreased. *C. heterochrous* was previously only found in European waters, including the coast of Denmark (Køie 2000a). Hansen & Malmstrøm (2006) was the first to find *C. heterochrous* in the inner Oslofjord (Midtmeie and Gråøyrenna), with American plaice and European plaice as the host. It was unknown for this nematode to be present in the inner Oslofjord until this research. Raamat (2012) also found *C. heterochrous* in the Oslofjord, this time at the middle part (Langøya). In my report I found some fish from the inner Oslofjord (Hellvik and Gråøyrenna) that were infected by *C. heterochrous,* this with the same hosts as found by Hansen & Malmstrøm (2006).

4.1 Methods

Some consideration should be mentioned regarding the sampling of fish. There was a different number of trawl hauls at the five locations, because of (1) a variable catch success of the flatfish at Drøbak and Gråøyrenna, and (2) some locations were chosen solely because of a larger abundances of flatfish (Midtmeie, Hellvik, and Tofteflaket). When comparing locations, a difference in number of trawl hauls may give an uneven result. Location with many hauls gives us a random distribution of the species, while a location with small samples sizes gives measurements which are not representable at that station.

Due to a large number of fish and time constrictions; most fish had to be frozen in the laboratory at the University of Oslo before dissection. This causes some issues because the best way to detect the nematode and record their position is to dissect the fish as soon as possible, since nematodes have a tendency to migrate after the host dies (Berland 1989, 2006). In order to prevent nematode migration, the fish were frozen as rapidly as possible after the catch. When dissecting the fish, I carefully inspect the stomach, intestine and faeces on a light table.

4.2 Nematode infestation

In all my samples from all investigated stations, 61 out of 383 individuals were infected by nematodes with an abundance of 0.5 worms per individual and an intensity of 3.5 worms per infected fish (Appendix II). There was a large geographical difference: in the middle Oslofjord (Tofteflaket and Drøbak) 44 of 83 investigated individuals were infected by nematodes, in the inner (Hellvik, Midtmeie and Gråøyrenna) only 17 of 300 were infected.

Hysterothylacium aduncum was the most abundant nematode specie in all sampled fish (14.9 % was infected by this nematode). *H.* aduncum was found at 4 of 5 stations with an intensity of 3.4. In contrast, *Cucullanus heterochrous* only infected 1 % of all sampled fish with an intensity of 5 per fish (Appendix II). This nematode was only observed at two stations (Hellvik and Gråøyrenna).

This high abundance of *H. aduncum* in my sample is consistent with other studies in the North Atlantic and the Oslofjord, where they all concluded that it is the most common and widespread nematode (Andersen 1993; Køie 1993; Balbuena *et* al. 1998; Raamat 2012). Only a few studies have reported that *C. heterochrous* is a nematode present in the Oslofjord (Jensen 1991; Hansen & Malmstrøm 2006; Raamat 2012).

In the middle of the Oslofjord, 74 % (Tofteflaket) and 27 % (Drøbak) of the sampled fish were infected by *H. aduncum*, whereas in the inner fjord the infections were only 5.4 % (Midtmeie) and 2.4 % (Hellvik). *C. heterochrous* was only found in the inner

Oslofjord (Hellvik and Gråøyrenna) where 3.6 % and 8.3 % respectively were infected by this nematode (Appendix II). In the middle Oslofjord the number of nematodes (all nematode species) per infected fish (all fish species) varied between 1.5 and 2.6. In the inner fjord the intensity varied between 0.05 and 0.9. This indicates that compared to the inner fjord, the nematodes are more present in the middle Oslofjord.

In the Oslofjord, also Hansen & Malmstrøm (2006) found a large prevalence of *H. aduncum* in the following fish species: Cod, Haddock, Whiting, American plaice, European plaice and Witch flounder. However, these were mostly found in the middle and outer Oslofjord. Raamat (2012) found more nematodes at a station located in the middle Oslofjord (Langøya) than in the outer (Torbjørnskjær). This indicates that *H. aduncum* is more abundant in the middle of Oslofjord, but still a large number is present in the outer.

Hysterothylacium aduncum matures in fish and cod is believed to be the final host (Berland 1961). The first intermediate hosts are various planktonic invertebrates (i.e. amphipods, copepods, isopods and shrimps; Berland 1991; Køie 1993; Klimpel & Rückert 2005). The amphipod *Themisto abyssorum* is the most important intermediate host for *H. aduncum* in the Oslofjord by Svendsen (1990). *Themisto abyssorum* is predated by many fish species. This is an important factor for the large abundances of *H. aduncum*'s in the fjord, due to the fact that many species of fish feed on it (Vinogradov 1999).

Cucullanus heterochrous was first time found in the inner Oslofjord in 2006 by Hansen & Malmstrøm. This nematode is mainly found in flounder and plaice (*Pleuronenectiformes*; Jensen 1991; Anderson 2000; Køie 2000a), and its larva has never been found in an invertebrate host. Køie (2000a) observed that polychaetes (*Nereis* spp., *Scolopos arminger*, *Brada villosa* and *Capitella* sp.) acted as intermediate hosts in her experimental infection in flatfish and cod. *Nereis diversicolor* seemed to be the most successful and important second intermediate of the polychaetes. Køie (1993) caught infected American plaice at depth of 45 m, whereas fish caught at depth of 342 m were not infected. A theory of her was that *N. diversicolor* is only naturally occurring down to 40 m (Køie 2000a). Hansen & Malmstrøm's (2006) trawled at depths between 80 and 100 m in the inner Oslofjord and 120 to 150 m in the outer Oslofjord. My trawling depths were 77 to 114 m in the inner Oslofjord, and 120 – 200 m in the middle.

The polychaetes mentioned in the previous section are not new in the fjord, and cannot explain why we found *C. heterochrous* in the inner Oslofjord. But since this nematode don't impose a treat to the fisheries, and thus no need for investigation; this specie may have been in the fjord in low abundances all along.

Codfish experienced a decrease of about 75 % during the last 20 years according to Hansen & Malmstrøm (2006), explaining it by the decline in the abundance of Atlantic cod in the Oslofjord (Svedäng & Bardon 2003). During the decline, flatfish may have then become a more available and suitable host, thus making it more infected. This raises the question whether flatfish has become a true second intermediate host in the Oslofjord.

4.2.1 American plaice

There was a great difference between the infection in American plaice in the inner Oslofjord (Hellvik and Midtmeie) where 15 of 272 investigated individuals were infected, and middle Oslofjord (Drøbak and Tofteflaket) where 39 of 66 were infected (Appendix II). The abundance in American plaice differed significantly between the middle and the inner part of the fjord. The level was about 51 times larger at Tofteflaket (3.05) than at Midtmeie (0.06; Table 3.2 a), and the intensity was 63 times larger at Drøbak than Midtmeie (1; Table 3.3 a).

Raamat (2012) investigated two stations; one in the middle (Langøya) and one in the outer Oslofjord (Torbjørnskjær) and reported that these two stations have an almost equal infection rate in this fish specie. Together with my results, this documents that American plaice is highly infected in the middle and outer Oslofjord, and may thus be considered as an important second intermediate host.

In my results 83 % of the 36 American plaice caught in Tofteflaket (middle Oslofjord) were infected by *H. aduncum*, the inner with 5.1 % and 6 % in Hellvik and Midtmeie respectively (Table 3.1 c). Hansen & Malmstrøm (2006) investigated fish from the inner, middle and outer Oslofjord and found only two infected American plaice (with *H. aduncum*). These were found in the inner and middle Oslofjord. But due to a small sample size, comparison is impossible. However, no previous investigations have reported that *H. aduncum* is present in American plaice; Aspholm (1991) for example found no *H. aduncum* in Torbjørnskjær. Raamat (2012) found that 44 % (of 116 investigated ind.) and 20 % (of 30 investigated ind.) of the American plaice were infected in Langøya and Torbjørnskjær respectively. Since the prevalence and abundance of *H. aduncum* also were at a high level in my samples, it seems that this nematode is on an increasing trend in the Oslofjord.

Cucullanus heterochrous in my samples was found in American plaice only at Hellvik (inner Oslofjord) where 2.5 % were infected. Of all investigated locations, the percentage of infected American plaice with *C. heterochrous* was only 0.6 % (Table 3.1 b). This is a rare nematode which previously only been found in the inner Oslofjord by Hansen & Malmstrøm (2006).

4.2.2 European plaice

In all samples only 23 individuals of European plaice were caught: 10 from the middle and 13 from the inner Oslofjord, where only 4 individuals were infected by nematodes (all nematode species). The percentage of infected European plaice (for all nematode species) was highest at three locations; two in the inner (Hellvik and Gråøyrenna) and one in the middle Oslofjord (Tofteflaket): 16 %, 100 % and 28 % respectively (Table 3.1 a).

The observation of *C. heterochrous* in the inner part of the fjord was unexpected, since only Hansen & Malmstrøm (2006) have observed this before. I found 13 individuals in European plaice from the inner Oslofjord (Appendix II). This is consistent with data from Hansen & Malmstrøm’s (2006) studies; 38 *C. heterochrous* was found in European plaice at Midtmeie and Gråøyrenna (inner Oslofjord).

They also found no *H. aduncum* present in European plaice. They trawled at different depths in the various part of the Oslofjord (80 - 100 m in the inner part and 120 – 150 m in the outer part). My catches were taken at roughly the same depths: 77-114 m in the inner and 120-200m in the middle. European plaice is widely distributed fish specie in depths less than 100 m in European waters (Hoarau *et* al. 2004).

The inner part of the Oslofjord clearly shows an increasing abundance of *C. heterochrous* and European plaice might be an adequate host for both of the two nematode species, but due to the low sample size a firm conclusion is not possible (Hansen & Malmstrøm 2006; Table 3.2 b).

4.2.3 Witch flounder

The percentage of infected Witch flounder (for all nematode species) was highest in the middle Oslofjord (Tofteflaket and Drøbak) with a prevalence of 66 % and 25 % respectively. There were no infected individuals from the inner part of the fjord (Table 3.1 a).

In my data set only 22 individuals was caught at all investigated stations, and of them 7 were caught at the middle Oslofjord (Table 3.1 a). At Drøbak the abundance was 0.5 among 4 individuals, all with *H. aduncum* (Table 3.2 c). No *C. heterochrous* was found in this fish specie. Due to the small sample size this result may be biased. Further, none of the 15 individuals from the inner Oslofjord was infected by nematodes.

Hansen & Malmstrøm (2006) found only 2 infected Witch flounder with 8 *H. aduncum* from the outer Oslofjord. However, their samples sizes were also low (a total of 15 individuals). Raamat (2012) found 4 infected individuals from the middle Oslofjord with 18 *H. aduncum.* But also her sample size was low (a total of 21 individuals).

These studies indicate that Witch flounder is a less important second intermediate host for nematodes. Witch flounders in the middle and the outer Oslofjord is more infected than Witch flounder in the inner fjord. *C. heterochrous* was not found in this fish specie. This might indicate that Witch flounder is not suitable as host for this nematode. However, the small sample sizes do not permit a firm conclusion.

4.2.4 *Hysterothylacium aduncum*

Hysterothylacium aduncum was found at all stations except Gråøyrenna. Of a total number of 57 identified nematodes in all fish species, 42 were found in American plaice. For this reason, the main focus in the analysis of my dataset will be on the infection by *H. aduncum* (Figure 3.2; Appendix II).

American plaice was mostly infected in the year classes 2 and 3 for both sexes (Figure I in Appendix IV). For the sexes individually, females had the highest abundances at the year classes 2 and 3 with respectively 13 and 10 individuals infected by *H. aduncum*. For males at the same year classes, there were 8 and 4 individuals infected (Figure 3.2). These were all sexually mature individuals. American plaice spawn between February and July with a peak in April and May. The dominance of infected American plaice in age group 2 and 3 might be because this age group is also the maturity age for the fish (O'brien *et* al. 1993); and thus gives a higher chance for these fish to be infected by nematodes because of the high abundances of mature fish at spawning time.

Figure 3.3 shows that Tofteflaket had the highest prevalence of infected American plaice, with 85 % of the females (a total of 68 nematodes) and 71 % of the males (a total of 26 nematodes) infected by *H. aduncum* (Table 3.7). Drøbak had the next highest infection rate, with 35 % of the females (a total of 54 nematodes) and 14 % of the males (a total of 3 nematodes) infected (Table 3.6). Both of these two stations are located in the middle Oslofjord. In comparison Midtmeie (inner Oslofjord) had a prevalence of 4 % in females (a total of 3 nematodes) and 7 % in males (a total of 7 nematodes). This station had the highest catch of 184 American plaice (Figure 3.3; Table 3.4). In Hellvik 78 individuals were caught, where only one female was infected (1 nematode; Table 3.5).

What this shows is that at all stations where American plaice were infected by *H. aduncum*, a higher percentage of females were generally infected (except at Midtmeie). This is in spite the fact that there was a larger catch of males than females. Raamat (2012) also investigated American plaice, but did not compare nematode infestation with the sex of the fish. However, when looking at her data set, it is clearly seen that females are more infected than males. While 45 females were infected with *H.* aduncum, only 6 males were infected. Andersen (1993) and Hansen & Malmstrøm (2006) also investigated *H. aduncum* in the Oslofjord, but did not mention the relationship between nematode infection and the sex of the fish.

The reason for the higher infection rate in females must necessarily be speculative, but according to Hoarau *et* al. (2004) a larger abundance of females is induced by a lower natural mortality. Solmundsson *et* al. (2003) studied plaice (*Pleuronectes* *platessa*) in Iceland and found that males tended to have a much higher recapture rate than females, especially during spawning season. The reason for this was that males have a different sexual behavior and spend more time in the spawning grounds than females. A possible explanation of the larger infection rate in females may therefore be that females are more abundant and move around in a larger area, so the probability of becoming infected is much larger than in males.

4.2.5 *Cucullanus heterochrous*

Cucullanus heterochrous was only found in American plaice and European plaice at two stations; Gråøyrenna and Hellvik (inner Oslofjord). 20 individuals were found in 4 fishes (11 in Gråøyrenna and 9 in Hellvik; Appendix II).

At Hellvik 78 American plaice were caught, with only one mature individual infected: a 2 year old male with 5 of this nematode (Figure 3.4). Also a juvenile was caught with two nematodes (Table 3.8). At Hellvik 6 European plaice were caught, but only a 3 year old male was infected with *Cucullanus* (2 ind*. Cucullanus*; Figure 3.7). At Gråøyrenna a single European plaice was caught. This was a 3 year old male with 11 nematodes inside (Figure 3.6 and 3.7).

This is consistent with the observations reported by Hansen & Malmstrøm (2006). They were the first to find *C. heterochrous* in the inner Oslofjord (Midtmeie and Gråøyrenna), within the same host as I did; American plaice and European plaice. The nematodes were distributed over 4 fish; one American plaice with one nematode and three European plaice with 38 nematodes. They also found a European flounder infected with 17 nematodes at Blåkollerenna (outer Oslofjord). Unfortunately they did not determine the fish age and sex of the nematode species. Raamat (2012) found one *C. heterochrous* in a female American plaice (3 year old) at Langøya in the middle part of the fjord.

4.3 Nematode length

The length of nematodes may give us an illustration of the hosts' species-specific adaptations. Ugland *et* al. (2004) showed that the average length of a nematode may be used as a measurement for the potential breeding in the final host, since the number of eggs (fecundity) is proportional to length. Since the body of the nematodes has different responses to the various fixation and identification fluids (Berland 1984), it is difficult to compare the measured length of the nematodes in the various published investigation.

4.3.1 *Hysterothylacium aduncum*

Hysterothylacium aduncum had the highest abundances at Tofteflaket (2.6; Appendix II). The average length of *H. aduncum* in all samples was 39.7 mm (Table 3.11). In the middle Oslofjord, Tofteflaket and Drøbak had an average length of 43.2 and 34.8 mm respectively. These two stations were also the ones with the highest number of nematodes present, 123 and 59 respectively (Appendix II). At Midtmeie and Hellvik (the inner Oslofjord) only 13 *H. aduncum* were found and these had a length between 20 and 30 mm. This suggests that *H. aduncum* are more abundant and have a larger size than in the middle Oslofjord.

In my investigation, *H. aduncum* infected American plaice (with 180 nematodes), European plaice (with 10 nematodes) and Witch flounder (5 with nematodes) and the corresponding average length were 37.6 mm, 69.8 mm and 51.6 mm respectively (Table 3.12; Appendix II). The nematodes obtained the shortest average length in American plaice, but they had the highest abundance in this fish species. Andersen (1993) found out that the same abundance of *H. aduncum* occurred for third-stage larvae in small and larger fish. This including with my results, may indicate that there is no differences in the infection rate when it comes to the size of the fish and that larger fish are not more suitable than smaller fish. Maturation of *H. aduncum* takes place inside the fish host, so length of the nematode is more dependent on the developmental stages (Berland 1961; Anderson 2000) rather than length of the fish host.

Hansen & Malmstrøm (2006) found 8 *H. aduncum* in Witch flounder (outer Oslofjord), but did not measure the length or recorded stages. Svendsen (1986) only found third-stage larvae in zooplankton samples from the outer Oslofjord (between Hvaler and Torbjørnskjær) with length from 0.9 mm to 16.6 mm. Klimpel & Rückert (2005) also found only L3 in hyperiid amphipods from the North Sea. These were identified as the obligatory intermediate host for this nematode, because fish foraging (Haddock and Whiting) in areas with large abundances of infected hyperiids showed a greater abundance of *H. aduncum*, whereas areas with scarce number of hyperiids showed fewer parasites in fish (Haddock and Whiting). Raamat (2012) reported that the L3 stage of *H. aduncum* had an average length of 16.0 mm in American plaice and Witch flounder. In my samples the average length was 18.7 mm in American plaice (no infected Witch flounder – Appendix III), and in both of our studies the highest abundance was found in the middle part of the fjord.

Gadoids are believed to be the final host (Berland 1961), and Andersen (1993) studied *H. aduncum* in cod from the outer Oslofjord (Tjøme), where the main goal was to see if there was any seasonality of infection during a year. The stage L3 was found to vary between 2.5 and 25.0 mm, L4 between 2.5 and 63.0 mm and L5 (adult worms) between 10.5 and 85.0 mm. In Raamat's (2012) samples, stage L3 varied between 10.0 – 23.0 mm, stage L4 between 10.0 – 61.0 mm and the adult stage between 17.0 – 66.0 mm. In my samples the data is roughly the same, L3 varied between 15.0 and 23.0 mm, L4 between 6.0 and 100.0 mm and the adult L5 worms measured between 16.0 to 67.0 mm (Table 4.1; Appendix III). In these studies the largest variability in size was found in L4, but the nematodes tended to be larger in my and Raamat's samples.

However the large difference to Andersen's (1993) results may be due to the fact that I only investigated flatfish. On the other hand, my results indicate that we should perhaps modify the commonly accepted viewpoint that gadoids are the main final host (Berland 1961; Andersen 1993). First, the larger size in the flatfish indicates that this group may be equally well suited as the final host. Secondly, the decline of codfish in the Oslofjord may have induced an increased infection in flatfish (Hansen & Malmstrøm 2006). But if so, that also is an indication that flatfish is a good substitute for codfish as a final host. Also, Køie (1993) stated that when *H. aduncum* larvae enter the fish, they have reached 2.0 – 3.0 mm. Because the large average sizes in L3 larvae reported by Raamat (2012) and me, flatfish should be considered a true second intermediate host.

Table 4.1. Length of *Hysterothylacium aduncum* stages L3, L4 and adults from three studies in the Oslofjord.

Hysterothylacium aduncum	**Third stage larvae**	**Fourth stage larvae**	**Adult worm**
Andersen (1993)	2.5 mm - 25 mm	2.5 mm - 63.0 mm	10.5 mm - 85.0 mm
Raamat (2012)	10.0 mm - 23.0 mm	10.0 mm - 61.0 mm	17.0 mm - 66.0 mm
Urskog (2014)	15.0 mm - 23.0 mm	6.0 mm - 100.0 mm	16.0 mm - 67.0 mm

4.3.2 *Cucullanus heterochrous*

Cucullanus heterochrous was found only at Gråøyrenna and Hellvik, with the highest abundance at the former station and an average length of 10.9 and 9.5 mm respectively (Appendix II). The average length of *C. heterochrous* in all the samples was 10.3 mm (Table 3.13).

This suggests that there are more *C. heterochrous* in inner part of the fjord, and that the size is almost equal at the two locations. Two fish species were infected, American plaice (5 individuals) and European plaice (13 individuals), where the average were 8.5 mm and 11.2 mm respectively (Table 3.14; Appendix II).

Køie (2000a) performed an experimental study where different intermediate hosts (invertebrates) were exposed to hundreds of infective eggs of *C. heterochrous* and then fed to flounders (*Platichthys flesus*) and plaice (*Pleuronectes platessa)*. These fish were considered to be the final host. All invertebrates were ingested, but only polyhaetes became infected, so Køie concluded that the polychaetes were the intermediate host. She further observed that third-stage larvae was about 550 µm to 1.1 mm one week after initial exposure, and moulted to fourth-stage when they reached about 800 µm to 1.4 mm. It is also likely that all or most *Cucullanus* spp. larvae emerging from eggs are third-stage, but this must be further investigated (Køie 2000 b). I only found L4 larvae and mature L5 worms, and they were all 7 mm or larger (Appendix III). This is consistent with her findings, when *C. heterochrous* is found in fish; they should be of a larger size and in fourth-stage or fifth-stage. The two largest *C. heterochrous* in my findings were 14 mm (L4 and male, both in European plaice).

Jensen (1991) found that parasites had a sexual dimorphism; 12.7 ± 0.8 mm for females and 8 ± 1.10 mm for males. These were caught in European flounder and European plaice at different locations in Norway, also some from the Oslofjord (Bunnefjord).

There is a general agreement that females have a larger average length than males in *C. heterochrous* (Berland 1970; Gibson 1972). However, only two female *C. heterochrous* were found in my data set (American plaice as the host); all shorter than males, 8.0 mm and 10.0 mm respectively. Some studies have been done in the fjord. Unfortunately Hansen & Malmstrøm (2006) did not measure the length of the parasites and also did not register the stages, which makes comparison impossible. Raamat (2012) also found this parasite in middle part of the fjord, a 12 mm long nematode with an unknown stage. The small sample sizes do not allow us to draw a conclusion.

4.4 Nematode length as a factor

Because of a large number of American plaice in the samples, the growth of *H. aduncum* was analyzed more detailed for the individuals occurring in this flatfish. A low number of *C. heterochrous* prevented a deeper analysis. Year classes 2 and 3 of both sexes of American plaice were the most infected. There was no relationship between the length of *H. aduncum* and the length or age of the host (Figure 3.8). The highest abundance of *H. aduncum* was observed in Tofteflaket (Figure 3.9).

Also Andersen (1993) did not find any correlation between fish size and worm length, i.e. the largest worms were found during the winter months independently of the fish size. Andersen further found the same abundances of *H. aduncum* third-stage larvae in small and larger fish. This indicates (1) large fish are not more suitable hosts, and (2) there is no preference for certain length group of fish.

4.5 Biological information of fish

4.5.1 Age distribution in American plaice

The most abundant fish species in all samples was American plaice (338 individuals). Most were caught at Midtmeie (inner Oslofjord) with a total of 184 individuals (Appendix II). There were slightly more males (94 ind.) than females (77 ind.), and the dominating year classes were 2 for females and 1 – 3 for males (Figure 3.10). At Hellvik (inner Oslofjord) the same patterns was observed (Figure 3.11).

Only 30 and 36 individuals of American plaice were caught at the two stations in the middle Oslofjord (Drøbak and Tofteflaket) respectively (Appendix II). In these samples more females were caught and the dominating age group for both sexes was age 2. Almost no 4 and 5 year old individuals were caught (Figure 3.12 and 3.13). This is quite peculiar because there seems to be a geographical difference of the caught males and females. Thus at all stations 2 and 3 year old fish was the most abundant.

According to Morgan & Colbourne (1999) American plaice at Newfoundland seemed to mature much earlier and have a relatively smaller size than earlier years (late 1960s to mid 1990s). Individuals in a cohort that had a lower average size during their lifespan had a higher fraction of individuals maturing at a younger age and smaller size. The males' average size from the Newfoundland area was 18 cm; this is a 28 % decrease from the late 1960s – early 1990s. The average sizes for females were 36 cm, a 10 % decrease from the same years.

They suggested that an increasing temperature may have influenced an earlier maturity, which for this species occur around 25 cm. On average, males mature earlier than females and at a smaller length (O'brien *et* al. 1993). In my samples there were quite few fish reaching over 30 cm (2 mature males in the age group 4 and 5), but these were all sexually mature individuals (Appendix I).

According to Thaulow & Faafeng (2013), the temperature in the air has increased the water temperature in the Oslofjord from 1930 – 2010. It is therefore likely that the sea temperature have influenced the biology of American plaice in the Oslofjord in the same way as in the Canadian waters.

In my data set there seemed to be more females over 20 cm (116 individuals) than males (71 individuals) at all locations. When looking at Raamat (2012) data set there was a dominance of females over 20 cm at her stations at the middle and outer Oslofjord (Referring to Raamat's Appendix I). Thus in our samples (obtained by trawling), individuals under 20 cm consist mostly of males. This is consistent with the fact that males tends to be smaller than females and as Morgan & Colbourne (1999) stated, also matures earlier.

4.5.2 Length vs age

The correlation between length and age was very variable for American plaice, between 27 % and 70 % at all stations (Figure 3.15). Since males tend to be smaller than females (Morgan & Colbourne 1999), and recruit to spawning grounds at an earlier age than females (Solmundsson *et* al. 2003); the general smaller sized younger males are expected to induce more variability in the correlation between length and age because of natural variation in growth rate at younger life stages.

4.5.3 Condition factor

According to Bolger & Connolly (1989) and Richter *et* al. (2000), fish in better condition is heavier at a given length. In my samples the length explained between 75 % and 99 % of the variability of weight (Figure 3.16), so there is a good covariation between weight and length in American plaice with a condition factor between 2.9 and 3.3. A condition factor under 3 indicates that larger individuals have changed their body shape to be more elongated or small individuals are in better condition. A condition factor over 3 indicates that larger individuals have increased their height or width more than length (Froese 2006). The low values of the condition factor (2.9) might be explained by the fact that flatfish are known to stop feeding and growing during winter time and therefore exhibit a seasonal change in the condition factor, with higher values in the summer than in the winter (Costopoulos & Fonds 1989).

4.6 Conclusions

Hysterothylacium aduncum was the most abundant nematode in all flatfish species at 4 of 5 locations with the highest abundance in the middle Oslofjord. Only a few individuals were infected by *H. aduncum* in the inner Oslofjord. *H. aduncum* can infect many planktonic invertebrates as intermediate hosts and subsequently use a wide range of fish species as final host.

Cucullanus heterochrous was only found in the inner Oslofjord. This nematode was previously found for the first time in the inner Oslofjord by Hansen & Malmstrøm (2006) and later Raamat (2012) also found this parasite in the middle Oslofjord

American plaice was infected by both nematode species, but mostly by *H.* aduncum. Two decades ago American plaice was not a known as a host in the fjord (Aspholm 1991), but during the last years this species has become an important host to the nematodes (Raamat 2012). My investigation confirms that American plaice is an important intermediate host.

European plaice was infected by both nematode species, but only a few individuals were infected.

For the second time *C. heterochrous* was found in the inner Oslofjord, and has so far only been observed in European plaice and American place.

Witch flounder was only found to be infected by a few *H. aduncums* and only in the middle Oslofjord. Hansen & Malmstrøm (2006) and Raamat (2012) also found only a few infected Witch flounder; so all these observations indicate that this fish is a less important second intermediate host.

American plaice mature at 2 and 3 years old, and these age groups were most infected (for all locations). Females were more infected than males. There was no correlation between the length of the nematodes and the condition of the host.

In the Oslofjord Sea temperature has increased during the last 80 years (Thaulow & Faafeng 2013), and this may have influenced the American plaice to mature at an earlier age and smaller size.

4.7 Future research

Hysterothylacium aduncum seems to be a “summer” species and mature individuals are more common in the warmer months (Berland 1998). Winter time introduce problems like low temperatures and limited food supply. This may force mature individuals to stop reproducing or leave site. In my samples large abundances of fourth stage larvae was found, but no seasonality was investigated. Future work should for this reason try to elaborate if there are more fourth and fifth stage larvae during the warmer months. Andersen (1993) found seasonality in cod of the different stages in the Oslofjord. To verify a seasonality various fish species should be sampled, especially flatfish because of their increasing infection rate. Also, investigate if this seasonality leads to more infected fish during these months.

Cod is believed to be the final host for *H. aduncum* (Berland 1961), but during the last two decades this fish species has decreased in the Oslofjord (Hansen & Malmstrøm 2006). In my samples a substantial number of *H. aduncum* was found in various flatfish species, this leads to the question whether flatfish has become a highly suitable intermediate host due to this decrease. Also, an interesting artifact is to investigate if the decrease of cod is an ongoing process in the Oslofjord.

The finding of *C. heterochrous* revealed that this specie is more common than first thought. Both my research and Hansen & Malmstrøm (2006) found this nematode to infect two flatfish species in the inner Oslofjord. Raamat (2012) also found some flatfish to be infected by this nematode in the middle Oslofjord. Future research should therefore try to find out if this nematode is spreading in the fjord.

American plaice in the year classes 2 and 3 was found in large abundances in my samples, these were all sexually mature individuals, and had a lower average length than expected. This leads to the question if increasing Sea temperature in the Oslofjord affects the biology of the flatfish to mature earlier and at smaller sizes (O’brien *et* al. 1993; Thaulow & Faafeng 2013).

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Personal comments

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APPENDIX

Appendix I

A complete list of all the fish dissected during 2013.

NR	FISH	GENDER	DATE	AREA	AGE	LENGTH (CM)	WEIGHT (GRAM)	H.a.	H.a.	H.a.	H.a.	C.h.	C.h.	C.h.	C.h.	Total
					(Years)			L3	L4	MALE	♀	L3	L4	Male	♀	
1	American plaice	FEMALE	21.02.2013	DRØBAK	3	22,5	99									0
2	American plaice	FEMALE	21.02.2013	DRØBAK	2	20,5	70									0
3	American plaice	FEMALE	21.02.2013	DRØBAK	3	22,5	76									0
4	American plaice	FEMALE	21.02.2013	DRØBAK	3	21,5	73		9	1						10
5	American plaice	FEMALE	21.02.2013	DRØBAK	3	26	141									0
6	American plaice	FEMALE	21.02.2013	DRØBAK	3	24	120									0
7	American plaice	FEMALE	21.02.2013	DRØBAK	3	23	102									0
8	American plaice	FEMALE	21.02.2013	DRØBAK	2	21,5	83		3							3
9	American plaice	FEMALE	21.02.2013	DRØBAK	2	23	83									0
10	American plaice	FEMALE	21.02.2013	DRØBAK	2	22,5	88		2							2
11	American plaice	FEMALE	21.02.2013	DRØBAK	3	22	75									0
12	American plaice	FEMALE	21.02.2013	DRØBAK	2	21	70									0
13	American plaice	FEMALE	21.02.2013	DRØBAK	2	22,5	90									0
14	American plaice	FEMALE	21.02.2013	DRØBAK	2	21	67		2							2
15	American plaice	MALE	21.02.2013	DRØBAK	1	19	40									0
16	American plaice	MALE	21.02.2013	DRØBAK	2	19	46									0
17	American plaice	MALE	21.02.2013	DRØBAK	2	21	80									0
18	American plaice	FEMALE	22.02.2013	DRØBAK	2	23	96									0
19	American plaice	FEMALE	22.02.2013	DRØBAK		22	93		3							3
20	American plaice	FEMALE	22.02.2013	DRØBAK	3	27	135									0
21	American plaice	FEMALE	22.02.2013	DRØBAK	2	23,5	95									0

22	American plaice	FEMALE	22.02.2013	DRØBAK	1	21	73	6	2				8
23	American plaice	FEMALE	22.02.2013	DRØBAK	1	19	61						0
24	American plaice	FEMALE	22.02.2013	DRØBAK	2	20	58	4					4
25	American plaice	FEMALE	22.02.2013	DRØBAK	2	20	49						0
26	American plaice	MALE	22.02.2013	DRØBAK	2	21,5	59						0
27	American plaice	MALE	22.02.2013	DRØBAK	2	20	54						0
28	American plaice	MALE	22.02.2013	DRØBAK	1	18	39	3					3
29	American plaice	MALE	22.02.2013	DRØBAK	2	18,5	44						0
30	American plaice	FEMALE	16.09.2013	DRØBAK	3	27	144,5	21		1			22
31	European plaice	FEMALE	16.09.2013	DRØBAK	3	36	590						0
32	European plaice	MALE	16.09.2013	DRØBAK		32,5	417,6						0
33	European plaice	MALE	16.09.2013	DRØBAK	3	30	310,3						0
34	Witch flounder		16.09.2013	DRØBAK	1	18	28,1						0
35	Witch flounder	FEMALE	16.09.2013	DRØBAK	3	27,5	135,1						0
36	Witch flounder	MALE	16.09.2013	DRØBAK	3	28	150,5	2					2
37	Witch flounder	MALE	16.09.2013	DRØBAK	2	29	167,7						0
38	American plaice	FEMALE	03.04.2013	GRÅØYRENNA	2	19,5	65,7						0
39	American plaice	MALE	03.04.2013	GRÅØYRENNA	1	18,5	40,3						0
40	American plaice	MALE	03.04.2013	GRÅØYRENNA	2	17	35,4						0
41	American plaice	MALE	03.04.2013	GRÅØYRENNA	2	22	74						0
42	American plaice	MALE	03.04.2013	GRÅØYRENNA	1	15,5	24,2						0
43	American plaice	MALE	03.04.2013	GRÅØYRENNA	2	19	53,6						0
44	American plaice	MALE	03.04.2013	GRÅØYRENNA	4	24,5	119,7						0
45	American plaice	MALE	03.04.2013	GRÅØYRENNA	4	32,5	316,4						0
46	American plaice	MALE	03.04.2013	GRÅØYRENNA	3	21	63,8						0
47	American plaice	MALE	03.04.2013	GRÅØYRENNA	2	20,5	56,9						0
48	European plaice	MALE	03.04.2013	GRÅØYRENNA	3	29,5	333,6				9	2	11
49	Witch flounder	FEMALE	03.04.2013	GRÅØYRENNA	3	34	344,4						0
50	American plaice		02.04.2013	HELLVIK	1	15,5	24,7						0
51	American plaice	FEMALE	02.04.2013	HELLVIK	3	23	102,5						0

52	American plaice	MALE	02.04.2013	HELLVIK	3	26	103,3						0
53	American plaice	MALE	02.04.2013	HELLVIK	3	25,5	90,3						0
54	American plaice	MALE	02.04.2013	HELLVIK	2	22	63			3	2		5
55	American plaice	MALE	02.04.2013	HELLVIK	2	20	48,4						0
56	American plaice	MALE	02.04.2013	HELLVIK	2	21	62,2						0
57	American plaice	MALE	02.04.2013	HELLVIK	2	17,5	33,9						0
58	American plaice	MALE	02.04.2013	HELLVIK	2	21	65						0
59	American plaice	MALE	02.04.2013	HELLVIK	3	23	92,9						0
60	American plaice	MALE	02.04.2013	HELLVIK	1	15,5	25,8						0
61	European flounder	MALE	02.04.2013	HELLVIK	2	31	373,5						0
62	European plaice	MALE	02.04.2013	HELLVIK	2	28	290,6						0
63	European plaice	FEMALE	02.04.2013	HELLVIK	2	28	276,8						0
64	European plaice	MALE	02.04.2013	HELLVIK	2	33	432,7						0
65	American plaice		26.06.2013	HELLVIK	1	19	62,6						0
66	American plaice		26.06.2013	HELLVIK	2	21	82,8					2	2
67	American plaice		26.06.2013	HELLVIK	1	18,5	50,5	1					1
68	American plaice		26.06.2013	HELLVIK	1	21,5	84						0
69	American plaice		26.06.2013	HELLVIK	1	17,5	42,1						0
70	American plaice		26.06.2013	HELLVIK	1	18,3	48,9						0
71	American plaice		26.06.2013	HELLVIK	1	18,2	52,8						0
72	American plaice		26.06.2013	HELLVIK	1	19	57,2						0
73	American plaice		26.06.2013	HELLVIK	1	12,4	13,5						0
74	American plaice	FEMALE	26.06.2013	HELLVIK	2	23	93,3		1				1
75	American plaice	FEMALE	26.06.2013	HELLVIK	2	25	108,7						0
76	American plaice	FEMALE	26.06.2013	HELLVIK	1	20,5	66						0
77	American plaice	FEMALE	26.06.2013	HELLVIK	1	20	63,6						0
78	American plaice	FEMALE	26.06.2013	HELLVIK	1	19,5	62,7						0
79	American plaice	FEMALE	26.06.2013	HELLVIK	1	19,5	57,5						0
80	American plaice	FEMALE	26.06.2013	HELLVIK	1	20	67						0
81	American plaice	FEMALE	26.06.2013	HELLVIK	2	21,5	70,5						0

82	American plaice	MALE	26.06.2013	HELLVIK	2	23	94	0
83	American plaice	MALE	26.06.2013	HELLVIK	2	19,5	67,6	0
84	American plaice	MALE	26.06.2013	HELLVIK	1	15,5	26,7	0
85	American plaice	MALE	26.06.2013	HELLVIK	1	18,1	45,8	0
86	American plaice	MALE	26.06.2013	HELLVIK	2	19	62	0
87	American plaice	MALE	26.06.2013	HELLVIK	2	21	88,5	0
88	American plaice	MALE	26.06.2013	HELLVIK	1	20	63,7	0
89	American plaice	MALE	26.06.2013	HELLVIK	1	17,5	51,4	0
90	American plaice	MALE	26.06.2013	HELLVIK	1	18	44,2	0
91	American plaice	MALE	26.06.2013	HELLVIK	2	17	42,9	0
92	American plaice	MALE	26.06.2013	HELLVIK	1	18,5	42,1	0
93	American plaice	MALE	26.06.2013	HELLVIK	2	16	35,7	0
94	American plaice	MALE	26.06.2013	HELLVIK	2	21,2	76,9	0
95	American plaice	MALE	26.06.2013	HELLVIK	2	20,5	65,1	0
96	American plaice	MALE	26.06.2013	HELLVIK	1	22	74,9	0
97	American plaice	MALE	26.06.2013	HELLVIK	2	19	52,7	0
98	American plaice	MALE	26.06.2013	HELLVIK	1	17	39,8	0
99	American plaice	MALE	26.06.2013	HELLVIK	1	19,5	47,7	0
100	American plaice	FEMALE	08.10.2013	HELLVIK	2	21,5	80,5	0
101	American plaice	FEMALE	08.10.2013	HELLVIK	2	21,5	71,5	0
102	American plaice	FEMALE	08.10.2013	HELLVIK	3	26	151,3	0
103	American plaice	FEMALE	08.10.2013	HELLVIK	4	28	178,7	0
104	American plaice	FEMALE	08.10.2013	HELLVIK	3	24,5	136,4	0
105	American plaice	FEMALE	08.10.2013	HELLVIK	2	23	108,2	0
106	American plaice	FEMALE	08.10.2013	HELLVIK	2	25,5	152,2	0
107	American plaice	FEMALE	08.10.2013	HELLVIK	3	24,5	136,4	0
108	American plaice	FEMALE	08.10.2013	HELLVIK	4	26	162,5	0
109	American plaice	MALE	08.10.2013	HELLVIK	1	14,5	24	0
110	American plaice	MALE	08.10.2013	HELLVIK	2	15	28	0
111	American plaice	MALE	08.10.2013	HELLVIK	2	18,5	54	0

112	American plaice	MALE	08.10.2013	HELLVIK	2	21	73,2		0
113	American plaice	MALE	08.10.2013	HELLVIK	1	14	22		0
114	American plaice	MALE	08.10.2013	HELLVIK	1	12	13		0
115	American plaice	MALE	08.10.2013	HELLVIK	1	13	18		0
116	American plaice	MALE	08.10.2013	HELLVIK	2	18	41		0
117	American plaice	MALE	08.10.2013	HELLVIK	3	24,5	144		0
118	American plaice	MALE	08.10.2013	HELLVIK	2	23	108		0
119	American plaice	MALE	08.10.2013	HELLVIK	3	27	151,7		0
120	American plaice	FEMALE	12.11.2013	HELLVIK	2	24	117,5		0
121	American plaice	FEMALE	12.11.2013	HELLVIK	2	23,5	103,5		0
122	American plaice	FEMALE	12.11.2013	HELLVIK	3	23	98,9		0
123	American plaice	FEMALE	12.11.2013	HELLVIK	2	23	114,1		0
124	American plaice	FEMALE	12.11.2013	HELLVIK	2	23	100		0
125	American plaice	FEMALE	12.11.2013	HELLVIK	3	22,5	106,2		0
126	American plaice	MALE	12.11.2013	HELLVIK	1	13	14,9		0
127	American plaice	MALE	12.11.2013	HELLVIK	2	19,5	63,3		0
128	American plaice	MALE	12.11.2013	HELLVIK	2	19	53,9		0
129	American plaice	MALE	12.11.2013	HELLVIK	1	15	24,2		0
130	American plaice	MALE	12.11.2013	HELLVIK	1	13	16,6		0
131	American plaice	MALE	12.11.2013	HELLVIK	1	18	51,7		0
132	European plaice	FEMALE	12.11.2013	HELLVIK	3	37	716,7		0
133	European plaice	FEMALE	12.11.2013	HELLVIK	4	44	1249		0
134	European plaice	MALE	12.11.2013	HELLVIK	3	38,5	648,4	2	2
135	American plaice		11.03.2013	MIDTMEIE	1	15,2	20,8		0
136	American plaice		11.03.2013	MIDTMEIE	2	17,2	145		0
137	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	24,5	116		0
138	American plaice	FEMALE	11.03.2013	MIDTMEIE	3	21,5	82,4		0
139	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	19	53,8		0
140	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	19,5	57,5		0
141	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	19,5	55,5		0

142	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	17,5	42,8		0
143	American plaice	FEMALE	11.03.2013	MIDTMEIE	3	21	64,4		0
144	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	20	58,1		0
145	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	20,5	72,9	1	1
146	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	20	57,3		0
147	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	19,5	52,1		0
148	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	19	51		0
149	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	20,5	55,7		0
150	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	21,5	70		0
151	American plaice	FEMALE	11.03.2013	MIDTMEIE	3	19,5	58		0
152	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	16,5	34,5		0
153	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	18,5	48,2		0
154	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	19,5	50,1		0
155	American plaice	FEMALE	11.03.2013	MIDTMEIE	3	20	60,5		0
156	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	19,5	48,5		0
157	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	20,5	68,5		0
158	American plaice	FEMALE	11.03.2013	MIDTMEIE	3	21	63,5		0
159	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	19	50		0
160	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	22,2	66,3		0
161	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	22,5	85,9		0
162	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	16	27,7		0
163	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	22	77,9		0
164	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	20	49,7		0
165	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	21,5	65,4		0
166	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	18	45,9		0
167	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	18,3	48,7		0
168	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	18,5	47,9		0
169	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	20,5	60,7		0
170	American plaice	FEMALE	11.03.2013	MIDTMEIE	1	19	49,5		0
171	American plaice	FEMALE	11.03.2013	MIDTMEIE	2	17,8	40,4		0

172	American plaice	MALE	11.03.2013	MIDTMEIE	1	20,5	54,9		0
173	American plaice	MALE	11.03.2013	MIDTMEIE	1	17,5	35,8		0
174	American plaice	MALE	11.03.2013	MIDTMEIE	2	18,5	47		0
175	American plaice	MALE	11.03.2013	MIDTMEIE	1	15,5	24,7		0
176	American plaice	MALE	11.03.2013	MIDTMEIE	1	16	27,8		0
177	American plaice	MALE	11.03.2013	MIDTMEIE	1	15,5	30,4		0
178	American plaice	MALE	11.03.2013	MIDTMEIE	1	19,5	50	1	1
179	American plaice	MALE	11.03.2013	MIDTMEIE	1	12,5	12,3		0
180	American plaice	MALE	11.03.2013	MIDTMEIE	1	17	31,5		0
181	American plaice	MALE	11.03.2013	MIDTMEIE	1	16,5	33,8		0
182	American plaice	MALE	11.03.2013	MIDTMEIE	1	14,5	21		0
183	American plaice	MALE	11.03.2013	MIDTMEIE	1	16,5	29		0
184	American plaice	MALE	11.03.2013	MIDTMEIE	3	18,5	56,3		0
185	American plaice	MALE	11.03.2013	MIDTMEIE	1	13,7	17,2		0
186	American plaice	MALE	11.03.2013	MIDTMEIE	1	15,5	26,1		0
187	American plaice	MALE	11.03.2013	MIDTMEIE	1	16,5	30,5		0
188	American plaice	MALE	11.03.2013	MIDTMEIE	1	18,4	39,5		0
189	American plaice	MALE	11.03.2013	MIDTMEIE	3	24,5	110,7		0
190	American plaice	MALE	11.03.2013	MIDTMEIE	2	21	62,9		0
191	American plaice	MALE	11.03.2013	MIDTMEIE	1	17,3	33,3		0
192	American plaice	MALE	11.03.2013	MIDTMEIE	2	16	32,6		0
193	American plaice	MALE	11.03.2013	MIDTMEIE	2	21,5	74,9		0
194	American plaice	MALE	11.03.2013	MIDTMEIE	1	21	67,2		0
195	American plaice	MALE	11.03.2013	MIDTMEIE	1	14,7	24,2		0
196	American plaice	MALE	11.03.2013	MIDTMEIE	1	14,5	23,3		0
197	American plaice	MALE	11.03.2013	MIDTMEIE	1	15,2	24	1	1
198	American plaice	MALE	11.03.2013	MIDTMEIE	1	14,7	22,3		0
199	American plaice	MALE	11.03.2013	MIDTMEIE	1	15,5	29,2		0
200	European plaice	MALE	11.03.2013	MIDTMEIE		39	1350		0
201	Witch flounder	MALE	11.03.2013	MIDTMEIE	2	21	68,3		0

202	American plaice		12.03.2013	MIDTMEIE	1	14,5	26,9		0
203	American plaice	FEMALE	12.03.2013	MIDTMEIE	3	21,5	84,1		0
204	American plaice	FEMALE	12.03.2013	MIDTMEIE	2	17	40,1		0
205	American plaice	FEMALE	12.03.2013	MIDTMEIE	3	21	75,3		0
206	American plaice	FEMALE	12.03.2013	MIDTMEIE	2	20,5	67,4		0
207	American plaice	FEMALE	12.03.2013	MIDTMEIE	2	20	76,8		0
208	American plaice	FEMALE	12.03.2013	MIDTMEIE	1	18	49,1		0
209	American plaice	FEMALE	12.03.2013	MIDTMEIE	2	22	71,6		0
210	American plaice	FEMALE	12.03.2013	MIDTMEIE	2	21	76,5		0
211	American plaice	MALE	12.03.2013	MIDTMEIE	2	18,5	49		0
212	Common dab	MALE	12.03.2013	MIDTMEIE	3	41	839,8		0
213	Witch flounder	FEMALE	12.03.2013	MIDTMEIE	3	34,5	291,1		0
214	Witch flounder	MALE	12.03.2013	MIDTMEIE	3	35	374,4		0
215	Witch flounder	MALE	12.03.2013	MIDTMEIE	2	29,5	187,2		0
216	American plaice	FEMALE	02.04.2013	MIDTMEIE	2	19	46,4		0
217	American plaice	MALE	02.04.2013	MIDTMEIE	2	16	25		0
218	American plaice	MALE	02.04.2013	MIDTMEIE	2	18,5	45,5		0
219	American plaice	MALE	02.04.2013	MIDTMEIE	1	17,5	31,7		0
220	American plaice	MALE	02.04.2013	MIDTMEIE	1	19	40,9		0
221	American plaice	MALE	02.04.2013	MIDTMEIE	1	16,5	27,8		0
222	American plaice	MALE	02.04.2013	MIDTMEIE	1	19,5	44,3		0
223	American plaice	MALE	02.04.2013	MIDTMEIE	3	21,5	62,2		0
224	American plaice	MALE	02.04.2013	MIDTMEIE	3	26	131,5		0
225	American plaice	MALE	02.04.2013	MIDTMEIE	1	18	37,7		0
226	American plaice	MALE	02.04.2013	MIDTMEIE	3	25	103,1		0
227	American plaice	MALE	02.04.2013	MIDTMEIE	3	20	49,9		0
228	American plaice	FEMALE	03.04.2013	MIDTMEIE	1	18	38,4		0
229	American plaice	FEMALE	03.04.2013	MIDTMEIE	3	22	90,5	1	1
230	American plaice	FEMALE	03.04.2013	MIDTMEIE	3	22	80,1		0
231	American plaice	FEMALE	03.04.2013	MIDTMEIE	3	24	93,5		0

232	American plaice	MALE	03.04.2013	MIDTMEIE	1	17	32,2			0
233	American plaice	MALE	03.04.2013	MIDTMEIE	4	24	87,3			0
234	American plaice	MALE	03.04.2013	MIDTMEIE	2	22,5	88,5			0
235	American plaice	MALE	03.04.2013	MIDTMEIE	3	23	76,7			0
236	American plaice	MALE	03.04.2013	MIDTMEIE	2	13	18,7			0
237	American plaice	MALE	03.04.2013	MIDTMEIE	4	25,5	110,1			0
238	American plaice	MALE	03.04.2013	MIDTMEIE	4	25	100,8			0
239	American plaice	MALE	03.04.2013	MIDTMEIE	3	21	63,9			0
240	Witch flounder	FEMALE	03.04.2013	MIDTMEIE	3	33	272,7			0
241	Witch flounder	MALE	03.04.2013	MIDTMEIE	3	32	200,4			0
242	Witch flounder	MALE	03.04.2013	MIDTMEIE	3	32,5	249,7			0
243	American plaice		23.04.2013	MIDTMEIE	2	15,5	24,3			0
244	American plaice		23.04.2013	MIDTMEIE	1	14,5	23,2	1		1
245	American plaice	FEMALE	23.04.2013	MIDTMEIE	2	21	50,4			0
246	American plaice	FEMALE	23.04.2013	MIDTMEIE	1	19,5	50,2			0
247	American plaice	FEMALE	23.04.2013	MIDTMEIE	3	21,5	66,3			0
248	American plaice	FEMALE	23.04.2013	MIDTMEIE	3	21	70,2			0
249	American plaice	FEMALE	23.04.2013	MIDTMEIE		21,5	67,6			0
250	American plaice	FEMALE	23.04.2013	MIDTMEIE	3	21	67,9			0
251	American plaice	MALE	23.04.2013	MIDTMEIE	1	18,5	38			0
252	American plaice	MALE	23.04.2013	MIDTMEIE	2	22	69			0
253	American plaice	MALE	23.04.2013	MIDTMEIE	2	21,5	70,4			0
254	American plaice	MALE	23.04.2013	MIDTMEIE	3	21	59,6			0
255	American plaice	MALE	23.04.2013	MIDTMEIE	2	22	67,3			0
256	American plaice	MALE	23.04.2013	MIDTMEIE	3	22,5	81,4			0
257	American plaice	MALE	23.04.2013	MIDTMEIE	1	15,5	24,8			0
258	American plaice	MALE	23.04.2013	MIDTMEIE	5	30,5	205,3		1	1
259	American plaice	MALE	23.04.2013	MIDTMEIE	3	21,5	72,1			0
260	American plaice	MALE	23.04.2013	MIDTMEIE	3	24	102,1			0
261	American plaice	MALE	23.04.2013	MIDTMEIE	2	22,5	73,5			0

262	American plaice	MALE	23.04.2013	MIDTMEIE	3	20,5	55,2		0
263	American plaice	MALE	23.04.2013	MIDTMEIE	3	20,5	60,8		0
264	American plaice	MALE	23.04.2013	MIDTMEIE	2	18,5	41,9		0
265	American plaice	MALE	23.04.2013	MIDTMEIE	2	19	43,9		0
266	American plaice	MALE	23.04.2013	MIDTMEIE	2	20	56,7		0
267	American plaice	MALE	23.04.2013	MIDTMEIE		25,5	135,7		0
268	American plaice	MALE	23.04.2013	MIDTMEIE	1	19,5	50		0
269	American plaice	MALE	23.04.2013	MIDTMEIE		18	39,7		0
270	American plaice	MALE	23.04.2013	MIDTMEIE	2	18	39,7		0
271	American plaice	MALE	23.04.2013	MIDTMEIE	3	22,5	90,7	1	1
272	American plaice	MALE	23.04.2013	MIDTMEIE	2	20	48,7		0
273	American plaice	MALE	23.04.2013	MIDTMEIE	2	19	52,1		0
274	American plaice	MALE	23.04.2013	MIDTMEIE	1	17,5	32,5		0
275	American plaice	MALE	23.04.2013	MIDTMEIE	3	19	36,7		0
276	American plaice	MALE	23.04.2013	MIDTMEIE	2	18	40,3		0
277	American plaice	MALE	23.04.2013	MIDTMEIE	3	21	62		0
278	American plaice	MALE	23.04.2013	MIDTMEIE	4	25	103,8		0
279	American plaice	MALE	23.04.2013	MIDTMEIE	3	21	62,9		0
280	Witch flounder	FEMALE	23.04.2013	MIDTMEIE	3	29,5	223,4		0
281	Witch flounder	MALE	23.04.2013	MIDTMEIE	2	20,5	51,7		0
282	Witch flounder	MALE	23.04.2013	MIDTMEIE	2	22,5	67,9		0
283	Witch flounder	MALE	23.04.2013	MIDTMEIE	1	19,5	35,2		0
284	American plaice		07.05.2013	MIDTMEIE	1	16	28		0
285	American plaice		07.05.2013	MIDTMEIE	1	18	38,8		0
286	American plaice	FEMALE	07.05.2013	MIDTMEIE	1	17,5	37		0
287	American plaice	FEMALE	07.05.2013	MIDTMEIE	2	23	104,1		0
288	American plaice	FEMALE	07.05.2013	MIDTMEIE	4	23	106,5		0
289	American plaice	FEMALE	07.05.2013	MIDTMEIE	3	23	64,2		0
290	American plaice	FEMALE	07.05.2013	MIDTMEIE	1	19,5	57,1		0
291	American plaice	MALE	07.05.2013	MIDTMEIE	4	24	84,6		0

292	American plaice	MALE	07.05.2013	MIDTMEIE	3	24,5	99,5	1	1
293	American plaice	MALE	07.05.2013	MIDTMEIE	1	18	47,8		0
294	American plaice	MALE	07.05.2013	MIDTMEIE	3	23	86,5		0
295	American plaice	MALE	07.05.2013	MIDTMEIE	1	20	60,3		0
296	Witch flounder	MALE	07.05.2013	MIDTMEIE	3	32	220		0
297	Witch flounder	MALE	07.05.2013	MIDTMEIE	3	33	294,5		0
298	American plaice	FEMALE	17.09.2013	MIDTMEIE	2	18,5	435		0
299	American plaice	FEMALE	17.09.2013	MIDTMEIE	2	19	50,5		0
300	American plaice	FEMALE	17.09.2013	MIDTMEIE	2	20	60,3		0
301	American plaice	FEMALE	17.09.2013	MIDTMEIE	4	29,5	250,3		0
302	American plaice	FEMALE	17.09.2013	MIDTMEIE	3	23	97,3		0
303	American plaice	FEMALE	17.09.2013	MIDTMEIE	3	23,5	87,4		0
304	American plaice	FEMALE	17.09.2013	MIDTMEIE	3	25	121,6		0
305	American plaice	FEMALE	17.09.2013	MIDTMEIE	4	23,5	90,1		0
306	American plaice	FEMALE	17.09.2013	MIDTMEIE	4	25	120,9	1	1
307	American plaice	FEMALE	17.09.2013	MIDTMEIE	3	21	66,8		0
308	American plaice	FEMALE	17.09.2013	MIDTMEIE	2	20,5	59,8		0
309	American plaice	FEMALE	17.09.2013	MIDTMEIE	2	20,5	61		0
310	American plaice	MALE	17.09.2013	MIDTMEIE	2	15	27,9		0
311	American plaice	MALE	17.09.2013	MIDTMEIE	2	17	37,7		0
312	American plaice	MALE	17.09.2013	MIDTMEIE	2	16,5	35,5		0
313	American plaice	MALE	17.09.2013	MIDTMEIE	2	15,5	24,4		0
314	American plaice	MALE	17.09.2013	MIDTMEIE	2	17,5	38,8		0
315	American plaice	MALE	17.09.2013	MIDTMEIE	2	15	20,7	1	1
316	American plaice	MALE	17.09.2013	MIDTMEIE	2	16,5	33	1	1
317	American plaice	MALE	17.09.2013	MIDTMEIE	4	25	124		0
318	European plaice	FEMALE	17.09.2013	MIDTMEIE	3	36	679,2		0
319	European plaice	FEMALE	17.09.2013	MIDTMEIE	3	44	1045,6		0
320	American plaice		08.10.2013	MIDTMEIE	1	12,5	13,7		0
321	American plaice		08.10.2013	MIDTMEIE	1	12	11,8		0

322	American plaice	FEMALE	08.10.2013	MIDTMEIE	3	24	98,7					0
323	American plaice	FEMALE	08.10.2013	MIDTMEIE	2	19	52,8					0
324	American plaice	FEMALE	08.10.2013	MIDTMEIE	2	21,5	69,2					0
325	American plaice	MALE	08.10.2013	MIDTMEIE	3	17	32,1					0
326	European plaice	MALE	08.10.2013	MIDTMEIE	3	31,5	346,7					0
327	Witch flounder	MALE	08.10.2013	MIDTMEIE	2	25,5	103					0
328	American plaice		12.11.2013	MIDTMEIE	2	18	35,5					0
329	American plaice	FEMALE	12.11.2013	MIDTMEIE	2	21,5	88,7					0
330	American plaice	FEMALE	12.11.2013	MIDTMEIE	2	19,5	58,1					0
331	American plaice	FEMALE	12.11.2013	MIDTMEIE	2	21	76,4					0
332	American plaice	FEMALE	12.11.2013	MIDTMEIE	2	21,5	66,3					0
333	American plaice	MALE	12.11.2013	MIDTMEIE	5	28,5	194,6					0
334	American plaice	MALE	12.11.2013	MIDTMEIE	5	29	236,7					0
335	American plaice	MALE	12.11.2013	MIDTMEIE	2	20,5	60,7					0
336	American plaice	MALE	12.11.2013	MIDTMEIE	3	21,5	77,8					0
337	American plaice	MALE	12.11.2013	MIDTMEIE	2	19,5	53,6					0
338	European plaice	FEMALE	12.11.2013	MIDTMEIE	4	40,5	1002					0
339	European plaice	FEMALE	12.11.2013	MIDTMEIE	4	43	1230					0
340	American plaice	FEMALE	07.10.2013	TOFTEFLAKET	3	23,5	107,2		3	2		5
341	American plaice	FEMALE	07.10.2013	TOFTEFLAKET	3	23	95,9		6	4		10
342	American plaice	FEMALE	07.10.2013	TOFTEFLAKET	3	25,5	143,4		8	1		9
343	American plaice	FEMALE	07.10.2013	TOFTEFLAKET	3	22	90		6	2		8
344	American plaice	FEMALE	07.10.2013	TOFTEFLAKET	2	20	70,2		2		1	3
345	American plaice	FEMALE	07.10.2013	TOFTEFLAKET	2	21,5	91,3	2				2
346	American plaice	FEMALE	07.10.2013	TOFTEFLAKET	2	21,5	83,9		7	1		8
347	American plaice	MALE	07.10.2013	TOFTEFLAKET	2	21	83,7		3	2		5
348	American plaice	MALE	07.10.2013	TOFTEFLAKET	3	19,5	61,9	1	1			2
349	European plaice	FEMALE	07.10.2013	TOFTEFLAKET	3	44	1026,7		7	2		9
350	European plaice	FEMALE	07.10.2013	TOFTEFLAKET	3	31,5	416,7					0
351	European plaice	MALE	07.10.2013	TOFTEFLAKET	3	34,5	562,1			1		1

352	European plaice	MALE	07.10.2013	TOFTEFLAKET	2	25,5	183,9					0
353	American plaice		11.11.2013	TOFTEFLAKET	1	14,5	22					0
354	American plaice	FEMALE	11.11.2013	TOFTEFLAKET	2	22,5	88,7		1			1
355	American plaice	FEMALE	11.11.2013	TOFTEFLAKET	3	27	159,5				1	1
356	American plaice	FEMALE	11.11.2013	TOFTEFLAKET	4	25,5	132,5					0
357	American plaice	FEMALE	11.11.2013	TOFTEFLAKET	3	25	131		2			2
358	American plaice	FEMALE	11.11.2013	TOFTEFLAKET	3	24,5	119,4					0
359	American plaice	FEMALE	11.11.2013	TOFTEFLAKET	2	21	88,2		1			1
360	American plaice	FEMALE	11.11.2013	TOFTEFLAKET	2	21,5	82,7		2			2
361	American plaice	MALE	11.11.2013	TOFTEFLAKET	4	25,5	100	1	7			8
362	American plaice	MALE	11.11.2013	TOFTEFLAKET	3	25	102		1			1
363	American plaice	MALE	11.11.2013	TOFTEFLAKET	2	16,5	36,3		2			2
364	American plaice	MALE	11.11.2013	TOFTEFLAKET	2	17	38		1			1
365	American plaice	MALE	11.11.2013	TOFTEFLAKET	2	19	53,3					0
366	American plaice	MALE	11.11.2013	TOFTEFLAKET	2	18	48,7		2			2
367	European plaice	MALE	11.11.2013	TOFTEFLAKET	3	27	181,5					0
368	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	4	25,5	123,9					0
369	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	2	22	84,4		2			2
370	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	3	22,5	100,8		2			2
371	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	3	24	108,9		1	4		5
372	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	3	23	89,9		3			3
373	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	3	24	104		3			3
374	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	3	26	150,4		8	2		10
375	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	3	23	119,8		5	1		6
376	American plaice	FEMALE	10.12.2013	TOFTEFLAKET	2	22,5	102,5		1			1
377	American plaice	MALE	10.12.2013	TOFTEFLAKET	2	16	37,8		3			3
378	American plaice	MALE	10.12.2013	TOFTEFLAKET	2	19,5	54,8		1			1
379	American plaice	MALE	10.12.2013	TOFTEFLAKET	2	15,5	26,8					0
380	American plaice	MALE	10.12.2013	TOFTEFLAKET	2	18,5	56,8		1			1
381	European plaice	MALE	10.12.2013	TOFTEFLAKET	2	29	234,8					0

382	European plaice	MALE	10.12.2013	TOFTEFLAKET	3	38	590,6		0
383	Lemon sole	MALE	10.12.2013	TOFTEFLAKET		30	281		0
384	Witch flounder	MALE	10.12.2013	TOFTEFLAKET	2	32	194,4	1	1
385	Witch flounder	MALE	10.12.2013	TOFTEFLAKET	3	34,5	288,8		0
386	Witch flounder	MALE	10.12.2013	TOFTEFLAKET	2	35,5	339,4	2	2

Appendix II

Nematode infection parameters in all fish at five study areas.

I Parameters of both nematode species

Study area	Specie	No. of fish	Infected fish	No. of nematodes	Prevalence (%)	Abundance	Intensity
Midtmeie	American plaice	184	11	11	6 %	0.06	1
	European plaice	6	0	0	0 %	0	0
	Witch flounder	14	0	0	0 %	0	0
	Total	**204**	**11**	**11**	**5.36 %**	**0.053**	**1**
Hellvik	American plaice	78	4	9	5.1 %	0.11	2.25
	European plaice	6	1	2	16 %	0.33	2
	Witch flounder	0	0	0	0 %	0	0
	Total	**84**	**5**	**11**	**5.88 %**	**0.13**	**2.2**
Drøbak	American plaice	30	9	57	30 %	1.9	6.33
	European plaice	3	0	0	0 %	0	0
	Witch flounder	4	1	2	25 %	0.5	2
	Total	**37**	**10**	**59**	**27 %**	**1.59**	**5.9**
Tofteflaket	American plaice	36	30	110	83 %	3.05	3.66
	European plaice	7	2	10	28.5 %	1.43	5
	Witch flounder	3	2	3	66 %	1	1,5
	Total	**46**	**34**	**123**	**74 %**	**2.6**	**3.62**
Gråøyrenna	American plaice	10	0	0	0 %	0	0
	European plaice	1	1	11	100 %	11	11
	Witch flounder	1	0	0	0 %	0	0
	Total	**12**	**1**	**11**	**8.33 %**	**0.91**	**11**
	Total	**383**	**61**	**215**	**16 %**	**0.56**	**3.52**

II Parameters of *Cucullanus heterochrous*

Study area	Specie	No. of fish	Infected fish	No. of nematodes	Prevalence (%)	Abundance	Intensity
Midtmeie	American plaice	184	0	0	0 %	0	0
	European plaice	6	0	0	0 %	0	0
	Witch flounder	14	0	0	0 %	0	0
	Total	**204**	**0**	**0**	**0 %**	**0**	**0**
Hellvik	American plaice	78	2	7	2.5 %	0.09	3.5
	European plaice	6	1	2	16.6 %	0.33	2
	Witch flounder	0	0	0	0 %	0	0
	Total	**84**	**3**	**9**	**3.57 %**	**0.1**	**3**
Drøbak	American plaice	30	0	0	0 %	0	0
	European plaice	3	0	0	0 %	0	0
	Witch flounder	4	0	0	0 %	0	0
	Total	**37**	**0**	**0**	**0 %**	**0**	**0**
Tofteflaket	American plaice	36	0	0	0 %	0	0
	European plaice	7	0	0	0 %	0	0
	Witch flounder	3	0	0	0 %	0	0
	Total	**46**	**0**	**0**	**0 %**	**0**	**0**
Gråøyrenna	American plaice	10	0	0	0 %	0	0
	European plaice	1	1	11	100 %	11	11
	Witch flounder	1	0	0	0 %	0	0
	Total	**12**	**1**	**11**	**8.33 %**	**0.91**	**11**
	Total	**383**	**4**	**20**	**1 %**	**0.052**	**5**

III Parameters of *Hysterothylacium aduncum*

Study area	Specie	No. Of fish	Infected fish	No. of nematodes	Prevalence (%)	Abundance	Intensity
Midtmeie	American plaice	184	11	11	6 %	0.06	1
	European plaice	6	0	0	0 %	0	0
	Witch flounder	14	0	0	0 %	0	0
	Total	**204**	**11**	**11**	**5.36 %**	**0.053**	**1**
Hellvik	American plaice	78	2	2	2.5 %	0.025	1
	European plaice	6	0	0	0 %	0	0
	Witch flounder	0	0	0	0 %	0	0
	Total	**84**	**2**	**2**	**2.4 %**	**0.02**	**1**
Drøbak	American plaice	30	9	57	30 %	1.9	6.33
	European plaice	3	0	0	0 %	0	0
	Witch flounder	4	1	2	25 %	0.5	2
	Total	**37**	**10**	**59**	**27 %**	**1.59**	**5.9**
Tofteflaket	American plaice	36	30	110	83 %	3.05	3.66
	European plaice	7	2	10	28.5 %	1.43	5
	Witch flounder	3	2	3	66 %	1	1.5
	Total	**46**	**34**	**123**	**74 %**	**2.6**	**3.62**
Gråøyrenna	American plaice	10	0	0	0 %	0	0
	European plaice	1	0	0	0 %	0	0
	Witch flounder	1	0	0	0 %	0	0
	Total	**12**	**0**	**0**	**0 %**	**0**	**0**
	Total	**383**	**57**	**195**	**14.8 %**	**0.51**	**3.4**

Appendix III

Length measurements of both nematode species sampled at Midtmeie, Hellvik, Gråøyrenna, Drøbak and Tofteflaket.

	Fish	Area	Nematode	Stage	Length (mm)
1	American plaice	HELLVIK	*C. heterochrous*	L4	7
2	American plaice	HELLVIK	*C. heterochrous*	L4	10
3	American plaice	HELLVIK	*C. heterochrous*	L4	10
4	American plaice	HELLVIK	*C. heterochrous*	FEMALE	8
5	American plaice	HELLVIK	*C. heterochrous*	FEMALE	10
6	American plaice	HELLVIK	*C. heterochrous*	MALE	7
7	American plaice	HELLVIK	*C. heterochrous*	MALE	8
8	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	10
9	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	13
10	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	13
11	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	9
12	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	11
13	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	12
14	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	8
15	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	9
16	European plaice	GRÅØYRENNA	*C. heterochrous*	L4	9
17	European plaice	GRÅØYRENNA	*C. heterochrous*	MALE	14
18	European plaice	GRÅØYRENNA	*C. heterochrous*	MALE	12
19	European plaice	HELLVIK	*C. heterochrous*	L4	14
20	European plaice	HELLVIK	*C. heterochrous*	L4	12
21	American plaice	MIDTMEIE	*H. aduncum*	L3	20
22	American plaice	TOFTEFLAKET	*H. aduncum*	L3	15
23	American plaice	TOFTEFLAKET	*H. aduncum*	L3	17
24	American plaice	TOFTEFLAKET	*H. aduncum*	L3	23
25	American plaice	DRØBAK	*H. aduncum*	L4	16
26	American plaice	DRØBAK	*H. aduncum*	L4	24
27	American plaice	DRØBAK	*H. aduncum*	L4	15
28	American plaice	DRØBAK	*H. aduncum*	L4	22
29	American plaice	DRØBAK	*H. aduncum*	L4	21
30	American plaice	DRØBAK	*H. aduncum*	L4	34
31	American plaice	DRØBAK	*H. aduncum*	L4	30
32	American plaice	DRØBAK	*H. aduncum*	L4	34
33	American plaice	DRØBAK	*H. aduncum*	L4	60
34	American plaice	DRØBAK	*H. aduncum*	L4	37
35	American plaice	DRØBAK	*H. aduncum*	L4	51
36	American plaice	DRØBAK	*H. aduncum*	L4	22
37	American plaice	DRØBAK	*H. aduncum*	L4	23
38	American plaice	DRØBAK	*H. aduncum*	L4	23
39	American plaice	DRØBAK	*H. aduncum*	L4	13

40	American plaice	DRØBAK	*H. aduncum*	L4	9
41	American plaice	DRØBAK	*H. aduncum*	L4	21
42	American plaice	DRØBAK	*H. aduncum*	L4	25
43	American plaice	DRØBAK	*H. aduncum*	L4	27
44	American plaice	DRØBAK	*H. aduncum*	L4	17
45	American plaice	DRØBAK	*H. aduncum*	L4	14
46	American plaice	DRØBAK	*H. aduncum*	L4	6
47	American plaice	DRØBAK	*H. aduncum*	L4	28
48	American plaice	DRØBAK	*H. aduncum*	L4	29
49	American plaice	DRØBAK	*H. aduncum*	L4	30
50	American plaice	DRØBAK	*H. aduncum*	L4	15
51	American plaice	DRØBAK	*H. aduncum*	L4	30
52	American plaice	DRØBAK	*H. aduncum*	L4	28
53	American plaice	DRØBAK	*H. aduncum*	L4	34
54	American plaice	DRØBAK	*H. aduncum*	L4	7
55	American plaice	DRØBAK	*H. aduncum*	L4	8
56	American plaice	DRØBAK	*H. aduncum*	L4	12
57	American plaice	DRØBAK	*H. aduncum*	L4	38
58	American plaice	DRØBAK	*H. aduncum*	L4	47
59	American plaice	DRØBAK	*H. aduncum*	L4	60
60	American plaice	DRØBAK	*H. aduncum*	L4	50
61	American plaice	DRØBAK	*H. aduncum*	L4	63
62	American plaice	DRØBAK	*H. aduncum*	L4	72
63	American plaice	DRØBAK	*H. aduncum*	L4	53
64	American plaice	DRØBAK	*H. aduncum*	L4	48
65	American plaice	DRØBAK	*H. aduncum*	L4	51
66	American plaice	DRØBAK	*H. aduncum*	L4	51
67	American plaice	DRØBAK	*H. aduncum*	L4	67
68	American plaice	DRØBAK	*H. aduncum*	L4	77
69	American plaice	DRØBAK	*H. aduncum*	L4	61
70	American plaice	DRØBAK	*H. aduncum*	L4	40
71	American plaice	DRØBAK	*H. aduncum*	L4	34
72	American plaice	DRØBAK	*H. aduncum*	L4	42
73	American plaice	DRØBAK	*H. aduncum*	L4	40
74	American plaice	DRØBAK	*H. aduncum*	L4	50
75	American plaice	DRØBAK	*H. aduncum*	L4	37
76	American plaice	DRØBAK	*H. aduncum*	L4	48
77	American plaice	DRØBAK	*H. aduncum*	L4	50
78	Witch flounder	DRØBAK	*H. aduncum*	L4	55
79	Witch flounder	DRØBAK	*H. aduncum*	L4	52
80	American plaice	HELLVIK	*H. aduncum*	L4	20
81	American plaice	MIDTMEIE	*H. aduncum*	L4	22
82	American plaice	MIDTMEIE	*H. aduncum*	L4	33
83	American plaice	MIDTMEIE	*H. aduncum*	L4	14
84	American plaice	MIDTMEIE	*H. aduncum*	L4	12
85	American plaice	MIDTMEIE	*H. aduncum*	L4	28

86	American plaice	MIDTMEIE	*H. aduncum*	L4	45
87	American plaice	MIDTMEIE	*H. aduncum*	L4	27
88	American plaice	MIDTMEIE	*H. aduncum*	L4	40
89	American plaice	MIDTMEIE	*H. aduncum*	L4	27
90	American plaice	TOFTEFLAKET	*H. aduncum*	L4	35
91	American plaice	TOFTEFLAKET	*H. aduncum*	L4	46
92	American plaice	TOFTEFLAKET	*H. aduncum*	L4	50
93	American plaice	TOFTEFLAKET	*H. aduncum*	L4	56
94	American plaice	TOFTEFLAKET	*H. aduncum*	L4	59
95	American plaice	TOFTEFLAKET	*H. aduncum*	L4	38
96	American plaice	TOFTEFLAKET	*H. aduncum*	L4	62
97	American plaice	TOFTEFLAKET	*H. aduncum*	L4	67
98	American plaice	TOFTEFLAKET	*H. aduncum*	L4	60
99	American plaice	TOFTEFLAKET	*H. aduncum*	L4	29
100	American plaice	TOFTEFLAKET	*H. aduncum*	L4	40
101	American plaice	TOFTEFLAKET	*H. aduncum*	L4	41
102	American plaice	TOFTEFLAKET	*H. aduncum*	L4	42
103	American plaice	TOFTEFLAKET	*H. aduncum*	L4	65
104	American plaice	TOFTEFLAKET	*H. aduncum*	L4	66
105	American plaice	TOFTEFLAKET	*H. aduncum*	L4	78
106	American plaice	TOFTEFLAKET	*H. aduncum*	L4	81
107	American plaice	TOFTEFLAKET	*H. aduncum*	L4	41
108	American plaice	TOFTEFLAKET	*H. aduncum*	L4	44
109	American plaice	TOFTEFLAKET	*H. aduncum*	L4	15
110	American plaice	TOFTEFLAKET	*H. aduncum*	L4	30
111	American plaice	TOFTEFLAKET	*H. aduncum*	L4	52
112	American plaice	TOFTEFLAKET	*H. aduncum*	L4	55
113	American plaice	TOFTEFLAKET	*H. aduncum*	L4	35
114	American plaice	TOFTEFLAKET	*H. aduncum*	L4	45
115	American plaice	TOFTEFLAKET	*H. aduncum*	L4	26
116	American plaice	TOFTEFLAKET	*H. aduncum*	L4	53
117	American plaice	TOFTEFLAKET	*H. aduncum*	L4	56
118	American plaice	TOFTEFLAKET	*H. aduncum*	L4	55
119	American plaice	TOFTEFLAKET	*H. aduncum*	L4	62
120	American plaice	TOFTEFLAKET	*H. aduncum*	L4	56
121	American plaice	TOFTEFLAKET	*H. aduncum*	L4	51
122	American plaice	TOFTEFLAKET	*H. aduncum*	L4	27
123	American plaice	TOFTEFLAKET	*H. aduncum*	L4	57
124	American plaice	TOFTEFLAKET	*H. aduncum*	L4	37
125	American plaice	TOFTEFLAKET	*H. aduncum*	L4	40
126	American plaice	TOFTEFLAKET	*H. aduncum*	L4	32
127	American plaice	TOFTEFLAKET	*H. aduncum*	L4	35
128	American plaice	TOFTEFLAKET	*H. aduncum*	L4	42
129	American plaice	TOFTEFLAKET	*H. aduncum*	L4	10
130	American plaice	TOFTEFLAKET	*H. aduncum*	L4	27
131	American plaice	TOFTEFLAKET	*H. aduncum*	L4	28

132	American plaice	TOFTEFLAKET	*H. aduncum*	L4	24
133	American plaice	TOFTEFLAKET	*H. aduncum*	L4	48
134	American plaice	TOFTEFLAKET	*H. aduncum*	L4	21
135	American plaice	TOFTEFLAKET	*H. aduncum*	L4	57
136	American plaice	TOFTEFLAKET	*H. aduncum*	L4	46
137	American plaice	TOFTEFLAKET	*H. aduncum*	L4	33
138	American plaice	TOFTEFLAKET	*H. aduncum*	L4	30
139	American plaice	TOFTEFLAKET	*H. aduncum*	L4	47
140	American plaice	TOFTEFLAKET	*H. aduncum*	L4	40
141	American plaice	TOFTEFLAKET	*H. aduncum*	L4	32
142	American plaice	TOFTEFLAKET	*H. aduncum*	L4	39
143	American plaice	TOFTEFLAKET	*H. aduncum*	L4	42
144	American plaice	TOFTEFLAKET	*H. aduncum*	L4	27
145	American plaice	TOFTEFLAKET	*H. aduncum*	L4	41
146	American plaice	TOFTEFLAKET	*H. aduncum*	L4	22
147	American plaice	TOFTEFLAKET	*H. aduncum*	L4	45
148	American plaice	TOFTEFLAKET	*H. aduncum*	L4	25
149	American plaice	TOFTEFLAKET	*H. aduncum*	L4	37
150	American plaice	TOFTEFLAKET	*H. aduncum*	L4	47
151	American plaice	TOFTEFLAKET	*H. aduncum*	L4	33
152	American plaice	TOFTEFLAKET	*H. aduncum*	L4	37
153	American plaice	TOFTEFLAKET	*H. aduncum*	L4	38
154	American plaice	TOFTEFLAKET	*H. aduncum*	L4	15
155	American plaice	TOFTEFLAKET	*H. aduncum*	L4	33
156	American plaice	TOFTEFLAKET	*H. aduncum*	L4	30
157	American plaice	TOFTEFLAKET	*H. aduncum*	L4	38
158	American plaice	TOFTEFLAKET	*H. aduncum*	L4	46
159	American plaice	TOFTEFLAKET	*H. aduncum*	L4	21
160	American plaice	TOFTEFLAKET	*H. aduncum*	L4	51
161	American plaice	TOFTEFLAKET	*H. aduncum*	L4	46
162	American plaice	TOFTEFLAKET	*H. aduncum*	L4	50
163	American plaice	TOFTEFLAKET	*H. aduncum*	L4	49
164	American plaice	TOFTEFLAKET	*H. aduncum*	L4	58
165	American plaice	TOFTEFLAKET	*H. aduncum*	L4	25
166	American plaice	TOFTEFLAKET	*H. aduncum*	L4	35
167	American plaice	TOFTEFLAKET	*H. aduncum*	L4	31
168	American plaice	TOFTEFLAKET	*H. aduncum*	L4	38
169	American plaice	TOFTEFLAKET	*H. aduncum*	L4	53
170	American plaice	TOFTEFLAKET	*H. aduncum*	L4	40
171	American plaice	TOFTEFLAKET	*H. aduncum*	L4	25
172	American plaice	TOFTEFLAKET	*H. aduncum*	L4	25
173	American plaice	TOFTEFLAKET	*H. aduncum*	L4	40
174	American plaice	TOFTEFLAKET	*H. aduncum*	L4	17
175	American plaice	TOFTEFLAKET	*H. aduncum*	L4	37
176	European plaice	TOFTEFLAKET	*H. aduncum*	L4	72
177	European plaice	TOFTEFLAKET	*H. aduncum*	L4	68

178	European plaice	TOFTEFLAKET	*H. aduncum*	L4	100
179	European plaice	TOFTEFLAKET	*H. aduncum*	L4	98
180	European plaice	TOFTEFLAKET	*H. aduncum*	L4	41
181	European plaice	TOFTEFLAKET	*H. aduncum*	L4	61
182	European plaice	TOFTEFLAKET	*H. aduncum*	L4	92
183	Witch flounder	TOFTEFLAKET	*H. aduncum*	L4	63
184	Witch flounder	TOFTEFLAKET	*H. aduncum*	L4	36
185	Witch flounder	TOFTEFLAKET	*H. aduncum*	L4	52
186	American plaice	DRØBAK	*H. aduncum*	FEMALE	38
187	American plaice	DRØBAK	*H. aduncum*	MALE	29
188	American plaice	DRØBAK	*H. aduncum*	MALE	16
189	American plaice	DRØBAK	*H. aduncum*	MALE	22
190	American plaice	HELLVIK	*H. aduncum*	MALE	20
191	American plaice	MIDTMEIE	*H. aduncum*	MALE	53
192	American plaice	TOFTEFLAKET	*H. aduncum*	FEMALE	30
193	American plaice	TOFTEFLAKET	*H. aduncum*	FEMALE	57
194	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	39
195	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	42
196	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	47
197	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	47
198	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	50
199	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	47
200	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	48
201	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	44
202	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	33
203	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	35
204	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	47
205	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	30
206	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	25
207	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	48
208	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	55
209	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	25
210	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	40
211	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	53
212	American plaice	TOFTEFLAKET	*H. aduncum*	MALE	35
213	European plaice	TOFTEFLAKET	*H. aduncum*	MALE	67
214	European plaice	TOFTEFLAKET	*H. aduncum*	MALE	56
215	European plaice	TOFTEFLAKET	*H. aduncum*	MALE	43

Appendix IV

Figures:

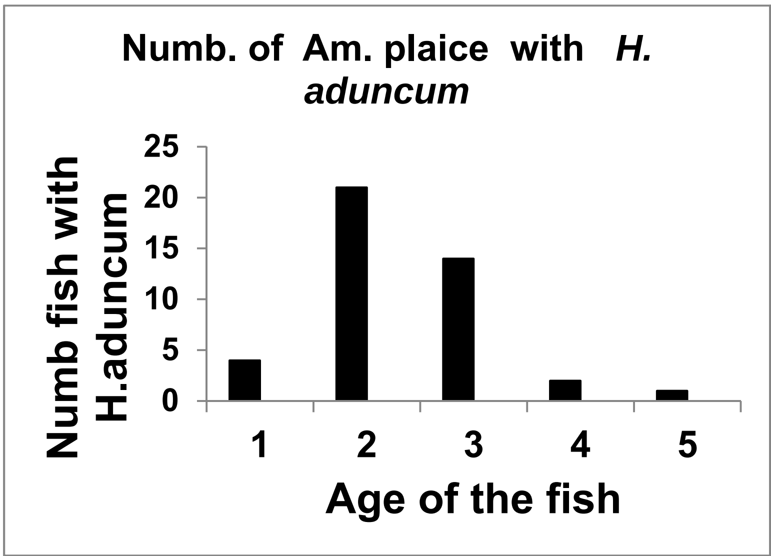

Figure I: Showing number of infected American plaice with *Hysterothylacium aduncum.* Age of the fish is shown at the x-axis.

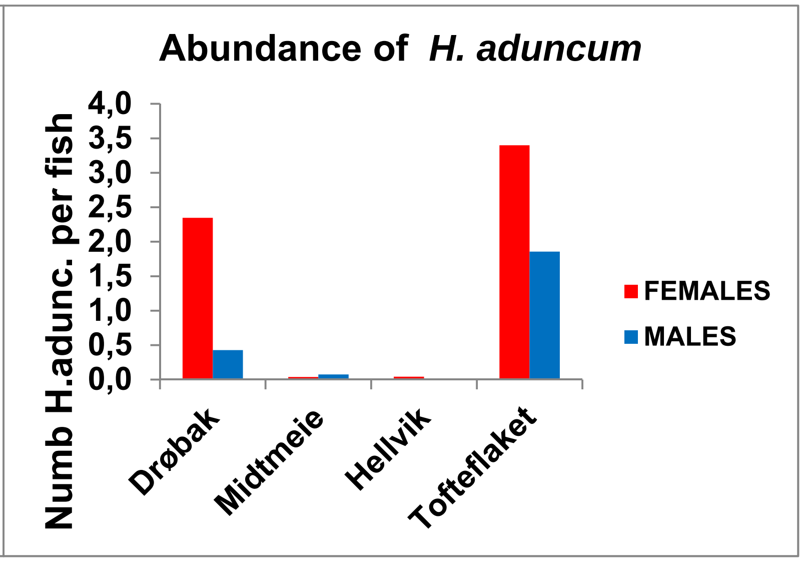

Figure II: Showing abundance of *Hysterothylacium aduncum* in females and males of American plaice at four stations. No *H. aduncum* was found at Gråøyrenna.

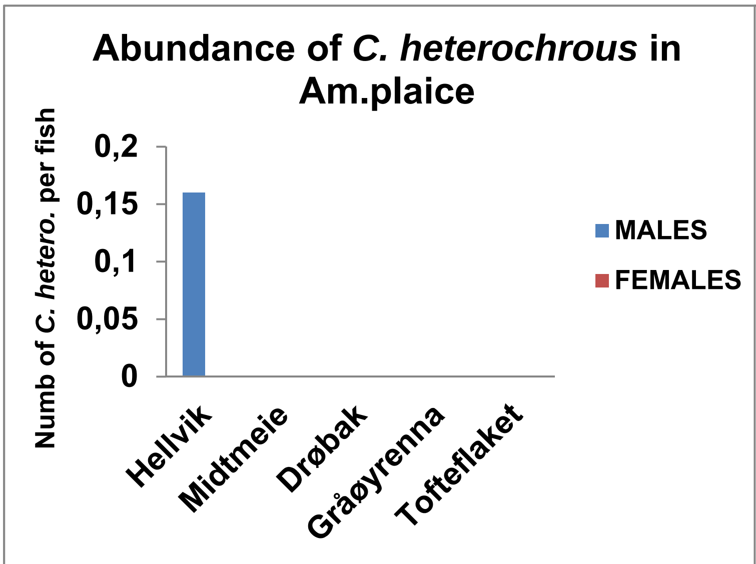

Figure III: Showing abundance of *Cucullanus heterochrous* in females and males of American plaice at five stations. Hellvik was the only location with *C. heterochrous*.

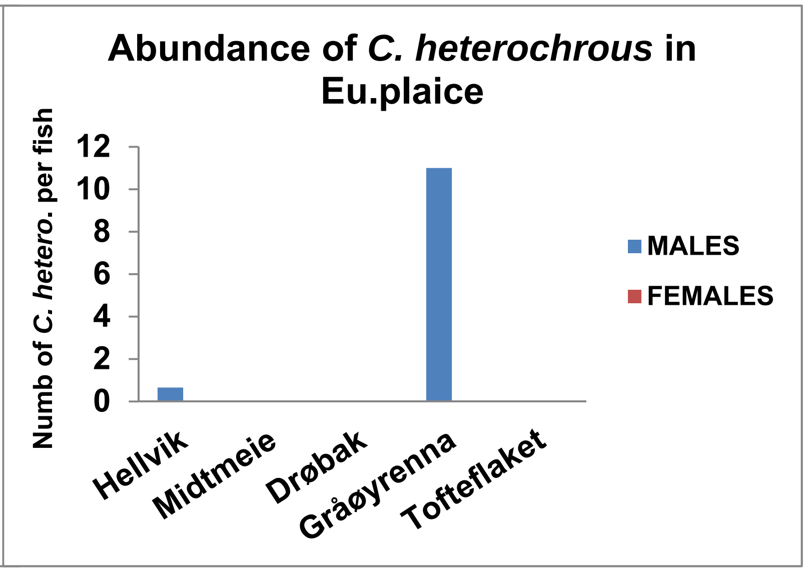

Figure IV: Showing abundance of *Cucullanus heterochrous* in females and males of European plaice at five stations. Hellvik and Gråøyrenna were the only locations with *C. heterochrous.*

Appendix V

Statistical calculations

Prevalence in female American plaice with respect to *Hysterothylacium aduncum* in the Oslofjord

4-sample test for equality of proportions without continuity correction

data: infish out of fish

X-squared = 71.8198, df = 3, p-value = 1.74e-15

alternative hypothesis: two.sided

sample estimates:

prop 1	prop 2	prop 3	prop 4
0.34782609	0.03896104	0.04166667	0.85000000

Conclusions: The prevalence of *H. aduncum* in female American plaice is significant different between the 4 investigated areas in the Oslofjord (P < 0.0001)

Prevalence in male American plaice with the respect to *Hysterothylacium aduncum* in the Oslofjord

4-sample test for equality of proportions without continuity correction

data: infish out of fish

X-squared = 57.0245, df = 3, p-value = 2.539e-12

alternative hypothesis: two.sided

sample estimates:

prop 1	prop 2	prop 3	prop 4
0.14285714	0.07446809	0.00000000	0.71428571

Conclusions: The prevalence of *H. aduncum* in male American plaice is significant different between the 4 investigated areas in the Oslofjord (P < 0.0001)

Age dependent infestation of *Hysterothylacium aduncum* in American plaice

Pearson's Chi-squared test with simulated p-value (based on 10000 replicates)

data: compUNIF

X-squared = 25.9904, df = NA, p-value = 9.999e-05

Conclusions: There is a significant difference in the prevalence of *H. aduncum* in the age groups of American plaice (P = 0.0001). The major infestation is found at age 2 and 3.

The length of *Hysterothylacium aduncum* versus the length of the host (American plaice)

From the excel file KIU 3 LENGTH DEPENDENCY sheets ALL STATIONS FEMALES & ALL STATIONS MALES & TOTAL SAMPLE we see that the variability in the length of the nematode is so large that it has no covariation with age and length of the host.

Female age distribution in the Oslofjord

Pearson's Chi-squared test with simulated p-value (based on 10000 replicates)

data: histf

X-squared = 11.6775, df = NA, p-value = 0.06959

Conclusions: The age distribution of females may be considered as significant different among the four areas (P = 0.070).

Male age distribution in the Oslofjord

Pearson's Chi-squared test with simulated p-value (based on 10000 replicates)

data: histf

X-squared = 20.3978, df = NA, p-value = 0.0019

Conclusions: The age distribution of males is significant different among the four areas (P = 0.002).

The additive model, females versus males at Midtmeie

area == "Midtmeie"

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	15.5873	0.4047	38.52	< 2e-16 ***
age	2.4011	0.1593	15.08	< 2e-16 ***
sexmales	-1.1566	0.3101	-3.73	0.000262 ***

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
age	1	924.20	924.20	227.02	< 2.2e-16 ***
sex	1	56.63	56.63	13.91	0.000262 ***
Residuals		168	683.92	4.07	

The interaction model

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	16.9330	0.6029	28.086	< 2e-16 ***
sexmales	-3.1931	0.7523	-4.245	3.62e-05 ***
sexfemales:age	1.7575	0.2675	6.569	6.16e-10 ***
sexmales:age	2.7307	0.1915	14.261	< 2e-16 ***

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
sex	1	55.49	55.49	14.261	0.000221 ***
sex:age	2	959.39	479.69	123.269	< 2.2e-16 ***
Residuals	167	649.87	3.89		

Comparison of models

Model 1: length ~ age + sex

Model 2: length ~ sex/age

	Res.Df	RSS	Df	Sum of Sq	F	Pr (>F)
1	168	683.92				
2	167	649.87	1	34.049	8.7498	0.003546 **

Conclusions: In Midtmeie the growth rate of females and males are significantly different (P = 0.004)

The additive model, females versus males at Hellvik

area == "Hellvik"

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	16.1906	0.9260	17.485	< 2e-16 ***
age	3.0741	0.3655	8.411	6.08e-12 ***
sexmales	-2.5768	0.5966	-4.319	5.56e-05 ***

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
age	1	522.49	522.49	105.648	3.534e-15 ***
sex	1	92.27	92.27	18.656	5.560e-05 ***
Residuals	64	316.52	4.95		

The interaction model

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	18.4165	1.1822	15.578	< 2e-16 ***
sexmales	-6.4142	1.4750	-4.349	5.10e-05 ***
sexfemales:age	2.0661	0.4984	4.145	0.000103 ***
sexmales:age	4.0233	0.4837	8.318	9.94e-12 ***

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
sex	1	264.84	264.845	59.358	1.174e-10 ***
sex:age	2	385.34	192.671	43.183	1.550e-12 ***
Residuals	63	281.09	4.462		

Comparison of models

Model 1: length ~ age + sex

Model 2: length ~ sex/age

	Res.Df	RSS	Df	Sum of Sq	F	Pr (>F)
1	64	316.52				
2	63	281.09	1	35.427	7.9401	0.006451 **

Conclusions: In Hellvik the growth rate of females and males are significant different (P = 0.006)

The additive model, females versus males at Drøbak

area == "Drøbak"

Coefficients

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	17.8254	1.1482	15.525	15.525 5.58e-15
age	2.0003	0.4785	4.180	0.00027
sexmales	-1.6831	0.7205	-2.336	0.027160

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
age	1	72.338	72.338	30.6798	7.187e-06
sex	1	12.867	12.867	5.4572	0.02716
Residuals	27	63.661	2.358		

The interaction model

Coefficients:

	Estimate	Std. Error	t value Pr (>|t|)
(Intercept)	17.6397	1.2498	14.114 1.06e-13 ***
sexmales	-0.6397	2.6294	-0.243 0.809690
sexfemales:age	2.0809	0.5237	3.973 0.000501 ***
sexmales:age	1.5000	1.3049	1.150 0.260812

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
sex	1	44.000	44.000	18.0881	0.0002411 ***
sex:age	2	41.620	20.810	8.5548	0.0013970 **
Residuals	26	63.246	2.433		

Comparison of models

Model 1: length ~ age + sex

Model 2: length ~ sex/age

	Res.Df	RSS	Df	Sum of Sq	F	Pr (>F)
1	27	63.661				
2	26	63.246	1	0.41517	0.1707	0.6829

Conclusion: In Drøbak the growth rate of females and males are not significant different (P = 0.683) but females are significant larger (P = 0.027). This means that either the females grow faster during the first two years or that larger females recruit the area.

The additive model, females versus males at Tofteflaket

area == "Tofteflaket"

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	14.1937	2.8324	5.011	2.08e-05 ***
age	3.3264	1.0039	3.314	0.00235 **
sexmales	-0.7488	1.3266	-0.564	0.57652

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
age	1	173.52	173.518	12.8358	0.001147 **
sex	1	4.31	4.307	0.3186	0.576519
Residuals	31	419.07	13.518		

The interaction model

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	17.396	3.491	4.983	2.44e-05 ***
sexmales	-8.348	5.191	-1.608	0.11825
sexfemales:age	2.140	1.258	1.701	0.09926
sexmales:age	5.192	1.578	3.291	0.00256 **

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
sex	1	29.40	29.395	2.2647	0.142806
sex:age	2	178.11	89.055	6.8612	0.003518 **
Residuals	30	389.38	12.979		

Comparison of models

Model 1: length ~ age + sex

Model 2: length ~ sex/age

	Res.Df	RSS	Df	Sum of Sq	F	Pr (>F)
1	31	419.07				
2	30	389.38	1	29.681	2.2868	0.1409

Conclusions: In Tofteflaket the growth rate of females and males are not significant different (P = 0.141) and the level of the growth curves for females and males are not significant different (P = 0.577). It is likely that the apparent equal growth rate of the sexes on Tofteflaket is due to the unusual large variability in the length at each age. The overall conclusion is that females grow faster than males.

The additive model, females versus area in the Oslofjord

sex == "females"

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	18.087140	0.487579	37.096	< 2e-16 ***
age	1.886713	0.160780	11.735	< 2e-16 ***
areaHellvik	0.725536	0.443835	1.635	0.104376
areaMidtmeie	-1.424293	0.362845	-3.925	0.000136 ***
areaTofteflaket	-0.006265	0.469100	-0.013	0.989364

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
age	1	397.63	397.63	172.075	< 2.2e-16 ***
area	3	107.29	35.76	15.477	9.658e-09 ***
Residuals	139	321.20	2.31		

The interaction model

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	17.6397	1.2270	14.376	< 2e-16 ***
areaHellvik	0.7768	1.4966	0.519	0.604596
areaMidtmeie	-0.7067	1.3132	-0.538	0.591336
areaTofteflaket	-0.2434	1.9254	-0.126	0.899603
areaDrøbak:age	2.0809	0.5141	4.047	8.65e-05 ***
areaHellvik:age	2.0661	0.3613	5.718	6.54e-08 ***
areaMidtmeie:age	1.7575	0.2077	8.463	3.68e-14 ***
areaTofteflaket:age	2.1402	0.5347	4.003	0.000102 ***

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
area	3	186.72	62.238	26.547	1.403e-13 ***
area:age	4	320.55	80.138	34.181	< 2.2e-16 ***
Residuals	136	318.85	2.345		

Comparison of models

Model 1: length ~ age + area

Model 2: length ~ area/age

	Res.Df	RSS	Df	Sum of Sq	F	Pr (>F)
1	139	321.20				
2	136	318.85	3	2.3468	0.3337	0.801

Conclusions: The growth rate of females are not significant different among the four areas (P = 0.801) but the level of the growth curves for females significant different between the subareas (P < 0.0001). This means that the size of the females are significant different between the studied subareas.

The additive model, males versus area in the Oslofjord

sex == "males"

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	14.4257	1.0998	13.116	<2e-16 ***
age	3.0017	0.2357	12.735	<2e-16 ***
areaHellvik	-0.6890	1.1031	-0.625	0.533
areaMidtmeie	-1.2537	1.0641	-1.178	0.241
areaTofteflaket	-0.2154	1.2620	-0.171	0.865

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
age	1	1226.32	1226.32	167.4249	<2e-16 ***
area	3	25.33	8.44	1.1529	0.3298
Residuals	153	1120.67	7.32		

The interaction model

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	17.0000	3.9517	4.302	3.04e-05 ***
areaHellvik	-4.9977	4.1054	-1.217	0.225
areaMidtmeie	-3.2601	3.9982	-0.815	0.416
areaTofteflaket	-7.9521	4.8669	-1.634	0.104
areaDrøbak:age	1.5000	2.2291	0.673	0.502
areaHellvik:age	4.0233	0.6101	6.594	6.85e-10 ***
areaMidtmeie:age	2.7307	0.2586	10.559	< 2e-16 ***
areaTofteflaket:age	5.1918	1.1668	4.450	1.67e-05 ***

Response: length

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
area	3	63.68	21.227	2.9904	0.03292 *
area:age	4	1243.90	310.976	43.8101	< 2e-16 ***
Residuals	150	1064.74	7.098		

Comparison of models

Model 1: length ~ age + area

Model 2: length ~ area/age

	Res.Df	RSS	Df	Sum of Sq	F	Pr (>F)
1	153	1120.7				
2	150	1064.7	3	55.927	2.6263	0.05253

Conclusions: The growth rate of males may be considered as significant different between the four areas (P = 0.053).

The additive model, female condition in the Oslofjord

sex == "females"

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	-4.97625	0.26990	-18.437	< 2e-16 ***
loglength	3.03308	0.08661	35.021	< 2e-16 ***
areaHellvik	0.07556	0.02942	2.568	0.01128 *
areaMidtmeie	-0.02988	0.02505	-1.193	0.23505
areaTofteflaket	0.09335	0.03089	3.023	0.00299 **

Response: logweight

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
loglength	1	18.5035	18.5035	1829.9027	< 2.2e-16 ***
area	3	0.2930	0.0977	9.6589	7.816e-06 ***
Residuals	139	1.4055	0.0101		

The interaction model

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	-4.4601	0.7228	-6.171	7.30e-09 ***
areaHellvik	-1.3339	0.9878	-1.350	0.179
areaMidtmeie	-0.6347	0.7952	-0.798	0.426
areaTofteflaket	1.0292	1.1601	0.887	0.377
areaDrøbak:loglength	2.8670	0.2325	12.330	< 2e-16 ***
areaHellvik:loglength	3.3183	0.2149	15.441	< 2e-16 ***
areaMidtmeie:loglength	3.0624	0.1098	27.901	< 2e-16 ***
areaTofteflaket:loglength	2.5708	0.2888	8.901	3.09e-15 ***

Response: logweight

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
area	3	6.3949	2.13163	213.70	< 2.2e-16 ***
area:loglength	4	12.4505	3.11263	312.04	< 2.2e-16 ***
Residuals	136	1.3566	0.00998		

Comparison of models

Model 1: logweight ~ loglength + area

Model 2: logweight ~ area/loglength

	Res.Df	RSS	Df	Sum of Sq	F	Pr (>F)
1	139	1.4055				
2	136	1.3566	3	0.048929	1.635	0.1842

Conclusions: The condition factor of females is not significant different among the four areas (P = 0.184) but the level of the condition factor for females is significant different between the subareas (P < 0.0001). This means that the level of condition at each age of the females are significant different between the studied subareas.

The additive model, male condition in the Oslofjord

sex == "males"

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	-5.03080	0.15369	-32.734	< 2e-16 ***
loglength	3.01085	0.04948	60.850	< 2e-16 ***
areaHellvik	0.13126	0.04833	2.716	0.007372 **
areaMidtmeie	0.02964	0.04640	0.639	0.523826
areaTofteflaket	0.18871	0.05487	3.439	0.000752 ***

Response: logweight

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
loglength	1	53.200	53.200	3795.098	< 2.2e-16 ***
area	3	0.546	0.182	12.983	1.336e-07 ***
Residuals	153	2.145	0.014		

The interactive model

Coefficients:

	Estimate	Std. Error	t value	Pr (>|t|)
(Intercept)	-5.92814	2.16735	-2.735	0.00699 **
areaHellvik	1.31430	2.18415	0.602	0.54825
areaMidtmeie	0.88350	2.17623	0.406	0.68534
areaTofteflaket	0.71244	2.20190	0.324	0.74673
areaDrøbak:loglength	3.31284	0.72924	4.543	1.13e-05 ***
areaHellvik:loglength	2.91288	0.09252	31.485	< 2e-16 ***
areaMidtmeie:loglength	3.02558	0.06640	45.565	< 2e-16 ***
areaTofteflaket:loglength	3.13439	0.12804	24.480	< 2e-16 ***

Response: logweight

	Df	Sum Sq	Mean Sq	F value	Pr (>F)
area	3	1.841	0.6138	43.58	< 2.2e-16 ***
area:loglength	4	51.936	12.9840	921.84	< 2.2e-16 ***
Residuals	150	2.113	0.0141		

Comparison of models

Model 1: logweight ~ loglength + area

Model 2: logweight ~ area/loglength

	Res.Df	RSS	Df	Sum of Sq	F	Pr (>F)
1	153	2.1448				
2	150	2.1127	3	0.032015	0.7577	0.5196

Conclusions: The condition factor of males is not significant different among the four areas (P = 0.520) but the level of the condition factor for males is significant different between the subareas (P < 0.0001). This means that the level of condition at each age of the males are significant different between the studied subareas.