Seasonal changes in the infection of young saithe, *Pollachius virens*, with *Anisakis simplex* and other helminths

Árstíðarbroytingar í rundorma-infektión í seiði

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Úrtak

Tilsamans 458 seiðir vórðu kannaðir við pepsin-saltsýru sodning fyri rundormar og aðrar sníkar í flaki og innvølum. Sýni vórðu tikin í Leirvíksfirði umleið annan hvønn mánaða frá januar til desember 1996. Fyrst í árinum var mest av 2-ára gomlum fiski, seinni á árinum var mest av 1-ára gomlum, og hendi hetta skifti so líðandi. Hjá 1-ára gomlum seiði var funnin týðuligur og stigvísur vøkstur við tíðini í títtleika (% infektión) av rundormunum *Anisakis simplex*, *Pseudoterranova decipiens* og *Contracaecum* sp. Fyri 2-ára gamlan seið vóru tølini eisini vaksandi fyrra partin av árinum men sýnast at náa einum meira jøvnum stigi seinni á árinum. Tølini geva ábendingar um ymiskan títtleika fyri ymsar árgangir av seiði. Miðal-tættleikin av teim trimum nevndu rundormunum (miðaltal av sníki í infiseraðum fiskum) var vaksandi fyri 1-ára gamlan seið, men tykist støðugt fyri 2-ára gamlan seið. Lutfalllið millum títtleikarnar av teim í stóran mun innkapslaðu rundormunum *Anisakis*, *Pseudoterranova* og *Contracaecum* broytist lítið fyri sama árgang av seiði. Aðrir funnir sníkar í innvølum vóru: rundormarnir *Hysterothylacium aduncum*, og *Cucculanus* sp. og tindormurin *Echinorhynchus gadi*. Vegna óstøðugar liviumstøður, í gørnunum serliga, eru títtleiki og tættleiki av hesum dýrum meira skiftandi og torgreiddir. Einstakt dømi var um krabbadýrið *Clavella* sp. og eitt plerocercoid av einum ónavngreindum bendilormi. Broytingarnar í *Anisakis*-infektión hjá upsa yvirhøvur tykjast at hava eitt øvugt samband við gongdini í gróðrinum í sjónum.

Abstract

Samples of saithe, *Pollachius virens*, were taken approximately every second month, from Jan to Dec 1996. A total of 458 saithe (age 1-2 years and 3 singles 3 years old) were examined by pepsin-digestion and dissection for nematodes in the fillets and the viscera. For the 1-year old saithe the prevalence of infection in fillets and viscera of *Anisakis simplex* third stage larvae was increasing with time (from 40 % in May to 71 % in Dec). For the 2-year old the figures ranged from 18 % Jan, to 84 % in October (80 % in December), indicating an increase in infection with age and different levels between different age classes. The intensity of infection was increasing with time for the 1-year old, but appeared stable for the 2-year old. Similar patterns were observed for the intensity and prevalence of infection of saithe with third stage larvae of *Pseudoterranova decipiens* and *Contracaecum* sp., but the figures were generally lower than for *A. simplex*. The predominantly encapsulated nematodes *Anisakis*, *Pseudoterranova* and *Contracaecum* had almost fixed ratios among themselves, which reflects their stable environment. Other parasites found in the viscera were the nematodes *Hysterothylacium aduncum* (larvae and adults), *Cucculanus* sp. and the acanthocehpalean *Echinorhynchus gadi*. They exhibited a more complex and varying pattern of abundance and prevalence of infection, reflecting their unstable environment primarily in the gut lumen. A single record was made of the crustacean *Clavella* sp. (Laernoidea) and an unidentified plerocercoid of a cestode. The changes of the *Anisakis*-infection in saithe seem to have an inverse relationship with the changes of the primary production.

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Introduction

Among the Faroese public as well as in other fish-consuming countries it is a general view that the infection of fishes with nematodes exhibits seasonal changes (pers. observations). It is well documented that prevalence of infection (percentage infected) and intensity of infection (mean number of parasites per infected fish) with *Anisakis simplex* third stage larvae is increasing with age for several fish species (Wootten, 1978; McClelland *et al.*,1990). Thorough work dealing with parasites of saithe like that of Scott (1985) or Polyanski (1966) did not examine the effect of seasonality in detail, however.

The investigations of fishes for nematodes usually are made in age-classes entering commercial fishery, leaving the younger age-classes relatively unexamined. Juvenile and young saithe is staying close to the coast, while the maturing age-classes do emigrate to deeper waters (Joensen and Tåning, 1970). This makes it difficult to catch juvenile or young fish with the common research vessels or commercial fishing vessels.

Infection of adult saithe from Faroese Waters with *A. simplex* in fillets was examined by Højgaard (1995a; b) and the present study is a continuation of this work. The aim was to search young saithe for parasites and to relate the infection with time of the year.

Materials and methods

Sampling

Young saithe was caught close to Leirvík, Faroe Islands from Jan to Dec, 1996 (Table 1). The sampling area is presumed to represent common Faroese bays and sounds, which are characterized by severe tidal currents, giving ideal conditions for young saithe to gather in schools and browse on the incoming zooplankton. The fish was caught at dawn with nylon longlines, operating from a traditional Faroese fishing boat. The distance to the coast was 50-300 m, bottom depth 10-60 m and fishing depth were approximately 1-7 m. Immediately after the sampling, the fish was weighed, the length was measured and the otoliths were removed. The whole fish were deep frozen in separate plastic bags at -18 °C.

Table 1. The samples of saithe from the Leirvík area, January-December 1996, positions close to 62°15' N, 06°40'W.
Talva 1. Tikin sýni av seiði úr Leirvíksfirði frá januar til desember 1996

Date	Number of saithe
05.01.96	28
26.01.96	47
17.02.96	45
23.03.96	75
25.05.96	47
24.08.96	77
26.10.96	78
14.12.96	33
27.12.96	28
Total	458

Digestion

After a thawing period of 6-12 h, butterfly fillets were cut from the saithe and the stomachs were refrozen. Digestion of the fillets without skin and the viscera in pepsin-hydrochloric acid were made in 2 L or 1/2 L beakers, respectively, at 37 °C for 3-6 h, pH 1-2 (Smith and Wootten, 1975). The fillets were digested with continuous

stirring on heating plates, the viscera (without the stomach) with occasional stirring in waterbaths. Adjustment of pH (with hydrochloric acid) was necessary after digestion in approx. half an hour, when the pH value could rise up to 4-6. The material digested were siewed through a 1mm mesh size. The nematodes found were identified in a microscope (40-100 x) or dissecting microscope (20 x). If the nematodes were too dark they were cleared in glycerol for 1-2 minutes.

Results

Samples of saithe

The 1- and 2-year old saithe seemed to follow a reciprocal immigration and emigration pattern. The 2-year old saithe dominated in the start of the year, but was gradually replaced by the 1-year old later on (Fig. 1). Saithe, 3-years old, was only found in 3 samples (all singles). The sex ratio was close to 1:1 for females and males (Fig. 2a,b). The growth expressed as an increase in length and weight appear to be most prominent from spring to autumn (Fig. 3a-d).

Anisakis simplex

The prevalence of infection increased steadily over the year for the 1-year old saithe (Fig. 4a). The same was found for the 2-year old saithe, but levelled out later in the year (winter) (Fig. 4b). The intensity of infection increased for the 1-year old saithe (Fig. 5a). However, no significant change in the intensity of infection for the 2-year old saithe could be seen (Fig. 5b). The intensity of infection for the 1-year old saithe is generally lower than for the 2-year old.

Other parasites found

Four other nematode species were frequently found: third-stage larvae of *Pseudoterranova decipiens* and *Contracaecum* sp. in the fillets and viscera, larvae and adults of *Hysterothylacium aduncum* and *Cucculanus* sp. in the viscera. The acanto-

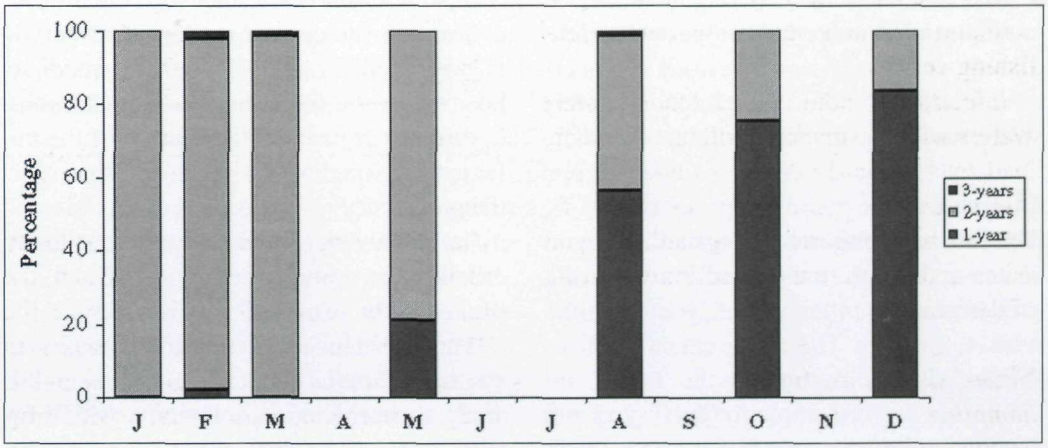

Fig. 1. *Age distribution of saithe samples from Leirvík, Faroe Islands, Jan-Dec. 1996 (n=458).*
Aldursbýtið av seiði úr Leirvíksfirði 1996

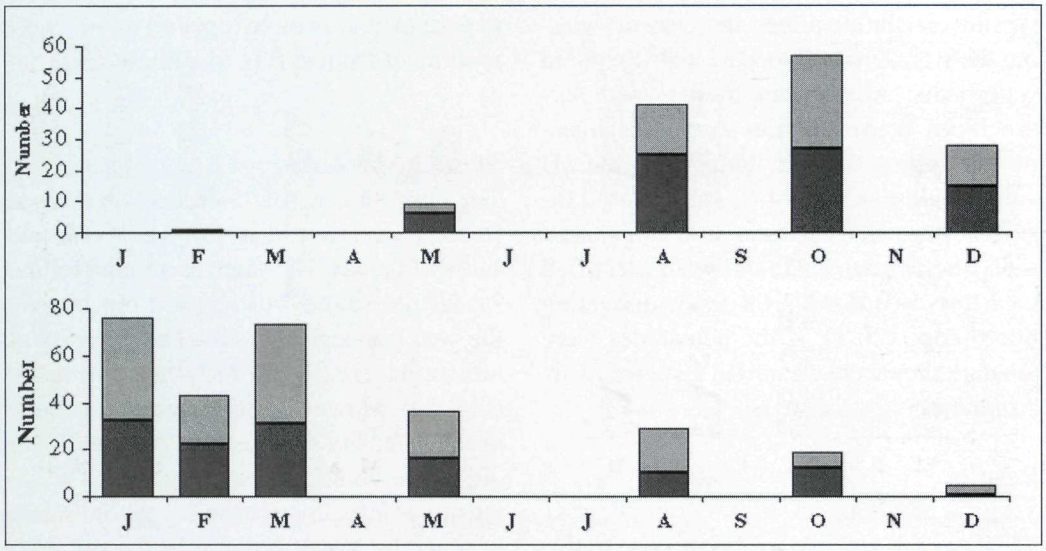

Fig. 2. a. The number of females (white) and males (black) in samples of 1-year old saithe.
Talið av kvennfiski (grátt) og kallfiski(svart) hjá 1-ára gomlum seiði.
Fig. 2. b. The number of females (white) and males (black) in samples of 2-year old saithe.
Talið av kvennfiski (grátt) og kallfiski (svart) hjá 2-ára gomlum seiði.

cephalean *Echinorhynchus gadi* were recovered from the gut lumen (adults) or in the pyloric caeca (larvae). The prevalence and intensity of infection are shown in Figs. 4 and 6. A single record was made of the crustacean laerneopodid ectoparasite *Clavella* sp. and an unknown cestode plerocercoid. As the stomach is not included in this study, no digeneans have been recorded.

Discussion

The results in this study showed a general increase through one calendar year in the prevalence of infection of young saithe with *A. simplex*. The mean intensity of infection was increasing for the 1-year old saithe, but seemed stable for the 2- year old saithe. Similar patterns, but at lower levels were revealed for *P. decipiens* and *Contracaecum* sp. How can these findings be interpreted?

A population of fishes which is infected by a population of parasites may be so in two principal ways. Either the infection is continuous (an even or random distribution is presumed for the infected intermediate host(s)), or the infection is non-continuous (a discrete or uneven distribution of the infected intermediate host). In the first case the prevalence of infection is expected to fit a simple linear function, but in the latter case a more complicated, non-linear function could be expected.

The prevalence of infection for *Anisakis*, *Pseudoterranova* and *Contracaecum* in this study appear to increase linearily with time (Fig. 4a), so the process of infection for saithe in this area most likely is continuous.

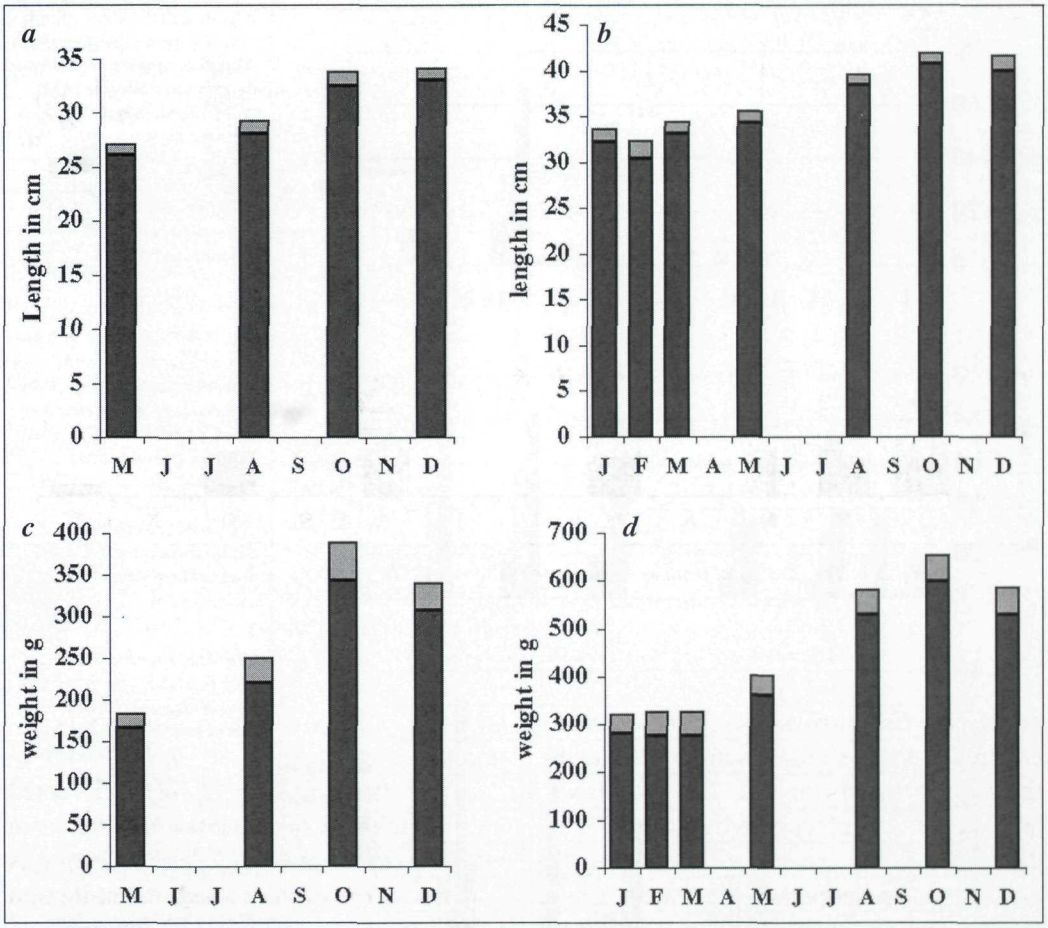

Fig. 3. a .Mean lengths (black) of 1-year old saithe; white = 0.5 S.D. (standard deviation)
Miðallongdir (svart) av 1-ára gomlum seiði; grátt = 0,5 standardfrávik.
Fig. 3. b. Mean lengths (black) of 2-year old saithe; white = 0.5 S.D
Miðallongdir (svart) av 2-ára gomlum seiði; grátt = 0,5 standardfrávik..
Fig. 3. c. Mean weights (black) of 1-year old saithe; white = 0.5 S.D.
Miðalvektir (svart) av 1-ára gomlum seiði; grátt = 0,5 standardfrávik.
Fig. 3. d. Mean weights (black) of 2-year old saithe; white = 0.5 S.D.
Miðalvektir (svart) av 2-ára gomlum seiði; grátt = 0,5 standardfrávik.

Højgaard (1995a) reported a general decline in the prevalence of infection and abundance (mean number of larvae per fish examined) with *A. simplex* in adult offshore saithe in the years 1991-1994. The present study includes younger fishes, 1- 2 years old. In this way the investigation reaches back to 1994 (the 1-year old fish has experienced 1995, and the 2-year old fish has experienced 1994 and 1995), thus connect-

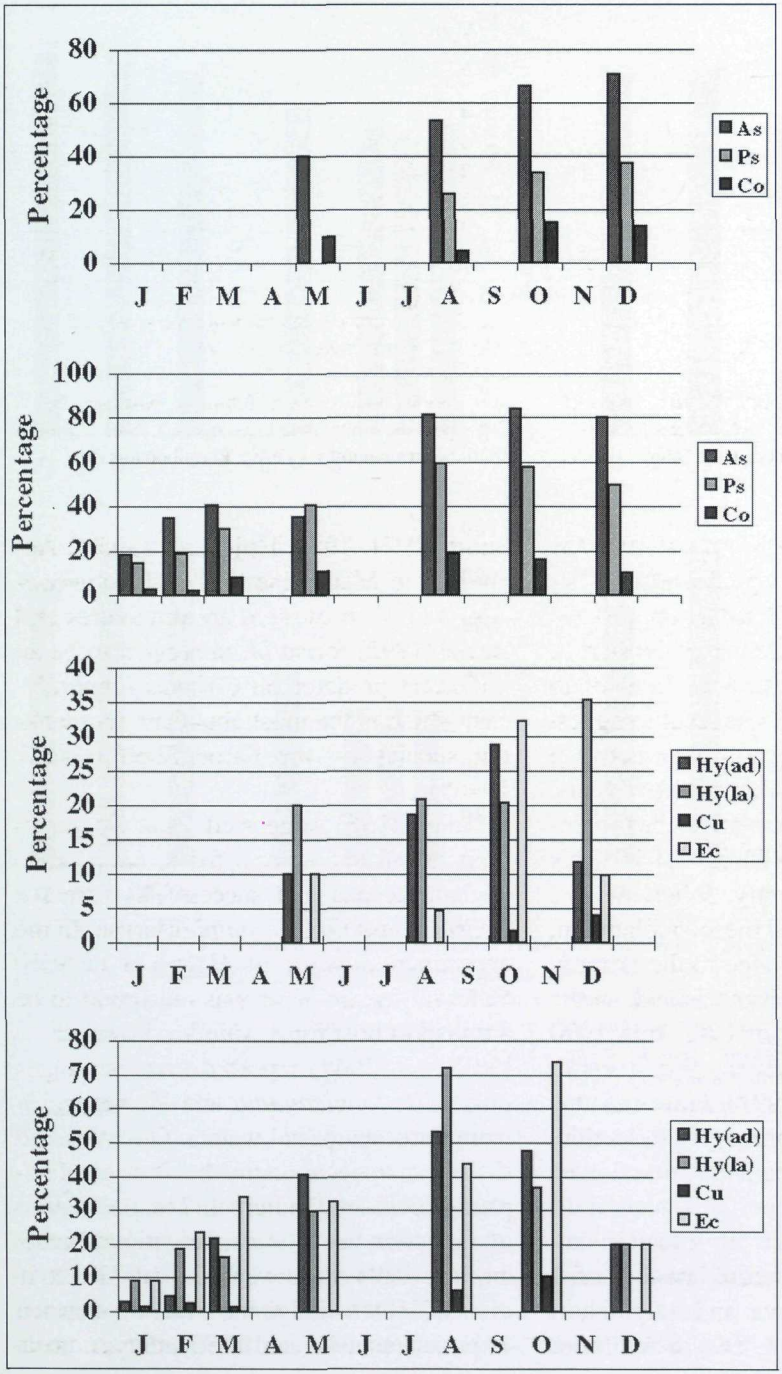

Fig. 4. a.*Prevalence of infection of 1-year old saithe with* ***Anisakis simplex*** *(As),* ***Pseudoterranova decipiens*** *(Ps) and* ***Contracaecum*** *sp. (Co).* Títtleiki av rundormasløgunum *Anisakis simplex* (As), *Pseudoterranova decipiens* (Ps) og *Contracaecum* sp. (Co) í 1-ára gomlum seiði.

Fig. 4. b.Prevalence of infection of 2-year old saithe with ***Anisakis simplex*** *(As),* ***Pseudoterranova decipiens*** *(Ps) and* ***Contracaecum*** *sp. (Co).* Títtleiki av rundormasløgunum *Anisakis simplex* (As), *Pseudoterranova decipiens* (Ps) og *Contracaecum* sp. (Co) í 2-ára gomlum seiði.

Fig. 4. c. *Prevalence of infection of 1-year old saithe with* ***Hysterothylacium aduncum****, adults=Hy(ad), larvae= Hy(la),* ***Cucculanus*** *sp. (Cu) and* ***Echinorhynchus gadi*** *(Ec).* Títtleiki av rundormasløgunum *Hysterothylacium aduncum* (Hy; ad = vaksin, la = larva), *Cucculanus sp.* (Cu) og tindormaslagnum *Echinorhynchus gadi* (Ec) í 1-ára gomlum seiði.

Fig. 4. d. *Prevalence of infection of 2-year old saithe with* ***Hysterothylacium aduncum****, adults=Hy(ad), larvae= Hy(la),* ***Cucculanus*** *sp. (Cu) and* ***Echinorhynchus gadi*** *(Ec).* Títtleiki av rundormasløgunum *Hysterothylacium aduncum* (Hy; ad = vaksin, la = larva), *Cucculanus sp.* (Cu) og tindormaslagnum *Echinorhynchus gadi* (Ec) í 1-ára gomlum seiði.

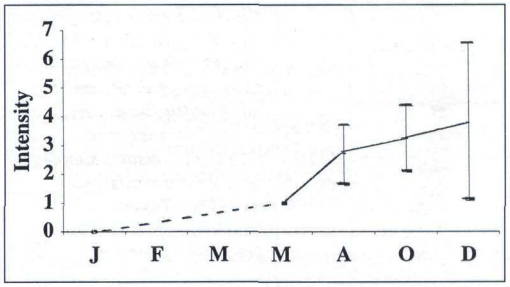

Fig. 5. a. Intensitiy of infection of 1-year old saithe with ***Anisakis simplex****; mean±S.D. (time scale is discontinous).*
Tættleiki av infektión av hvalormi, *Anisakis simplex*, í 1-ára gomlum seiði. Tølini eru meðaltøl ± standard-frávik; allir mánaðir í ári eru ikki umboðaðir.

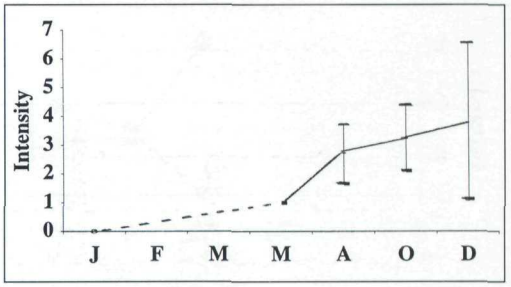

Fig. 5. b. Intensitiy of infection of 2-year old saithe with ***Anisakis simplex****; mean±S.D. (time scale is discontinous).*
Tættleiki av infektión av hvalormi, *Anisakis simplex*, í 2-ára gomlum seiði. Tølini eru meðaltøl ± standard-frávik; allir mánaðir í ári eru ikki umboðaðir.

ing the two studies. At the end of 1996 the 1-year old saithe had significantly higher levels of prevalence of infection (70 %) than the 2-year old saithe had at the start of the year (from 18 to 40% in January to March) (Figs. 4 a, b). These results suggest that the levels of infection vary between different age classes from different years. But they also indicate a relative change (increase) in the years 1995 and 1996. This observation is apparently following the changes in abundance of mesozooplankton, which was inversely related to the primary production around the Faroe Islands in the same period: a steady increase from 1990 to 1995, but much lower in 1996. (Gaard, 1996a; Gaard *et al.*, 1997). How can this correlation between primary production, mesozooplankton and parasitic infection be explained?

The intermediate hosts for *Anisakis simplex* are the euphausiaceans *Thysanoessa inermis*, *T. longicaudata* and *Meganyctiphanes norvegica* (see eg. Polyanski, 1966; Smith, 1971, 1983; Højgaard, 1995b). According to Mauchline (1980) *T. longicaudata* and *M. norvegica* are omnivores and Beyer (1992) found *M. norvegica* to be an important predator on *Calanus finmarchicus*, which is the most abundant zooplankton species on the Faroe Shelf (Gaard, 1996b).

Køie (1993) suggested calanoid copepods as transport hosts for *A. simplex*, to euphausiaceans and successfully infected *Acartia tonsa* with *A. simplex* larvae. In the experimental work of Højgaard (1995b) *Calanus finmarchicus* was not found to be a transport host for *A. simplex*, however. Einarsson (1945) reported juvenile stages only of *T. longicaudata* and *M. norvegica* from Faroese coastal waters. Otherwise no data seem to occur on the distribution of euphausiaceans at the Faroes. The prevalence of infection with *Anisakis* in euphausiaceans usally is low (Smith, 1983). It is thus difficult to judge the exact importance of the euphausiacean intermedatiate hosts

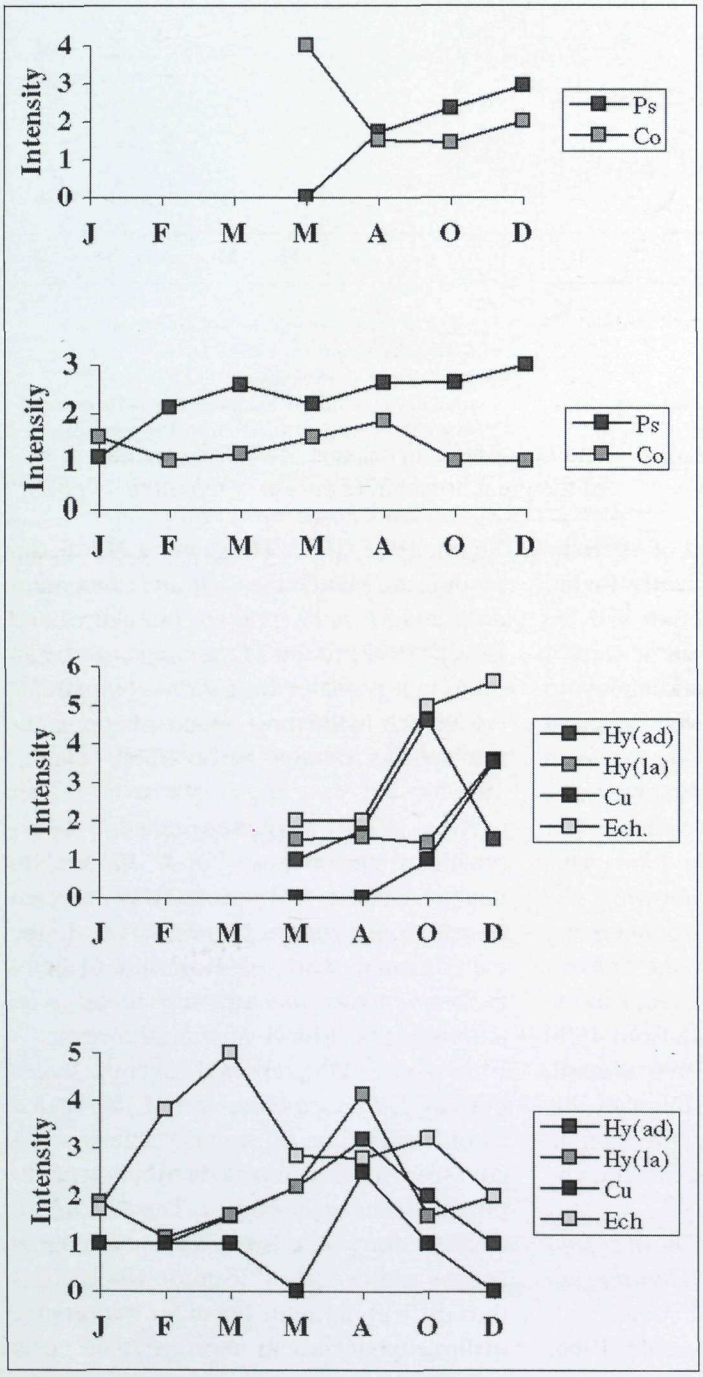

Fig. 6. a. *Intensity of infection of 1-year old saithe with* ***Pseudoterranova decipiens*** *(Ps) and* ***Contracaecum*** *sp. (Co).* Tættleiki av rundormasløgunum *Pseudoterranova decipiens* (Ps) og *Contracaecum* sp. (Co) í 1-ára gomlum seiði.

Fig. 6. b. *Intensity of infection of 2-year old saithe with* ***Pseudoterranova decipiens*** *(Ps) and* ***Contracaecum*** *sp. (Co).* Tættleiki av rundormasløgunum *Pseudoterranova decipiens* (Ps) og *Contracaecum* sp. (Co) í 2-ára gomlum seiði.

Fig. 6. c. *Intensity of infection of 1-year old saithe with* ***Hysterothylacium aduncum*** *adults (Hy(ad)) and larvae (Hy(la)),* ***Cucculanus*** *sp. (Cu) and* ***Echinorhynchus gadi*** *(Ec).* Tættleiki av rundormasløgunum *Hysterothylacium aduncum* (Hy; ad=vaksin, la=larva), *Cucculanus sp.* (Cu) og tindormaslagnum *Echinorhynchus gadi* (Ec) í 1-ára gomlum seiði.

Fig. 6 d. *Intensity of infection of 2-year old saithe with* ***Hysterothylacium aduncum*** *adults (Hy(ad)) and larvae (Hy(la)),* ***Cucculanus*** *sp. (Cu) and* ***Echinorhynchus gadi*** *(Ec).* Tættleiki av rundormasløgunum *Hysterothylacium aduncum* (Hy; ad=vaksin, la=larva), *Cucculanus sp.* (Cu) og tindormaslagnum *Echinorhynchus gadi* (Ec) í 2-ára gomlum seiði.

of *A. simplex*, but the general biology and the population dynamics of the intermediate hosts are of obvious importance. If the population of the euphausiacean intermeditae host are at low level the possibility also will be low for the *Anisakis* larvae to reach the saithe through infected euphausiacean. If the euphausiacean population are at a high level it will become more likely for the saithe to be infected by the *Anisakis* larvae. The preconditions for this assumption is a steady supply of *Anisakis*-eggs from the final host, the whales, and subsequent hatching of larvae and infection of the euphausiaceans. Due to the high turbulence in the water on the Faroese Shelf, *Anisakis*-eggs and larvae must be assumed to follow the same patterns of total mixing as do the nutrients (Hansen, 1992).

As the long finned pilot whale, *Globicephala melas*, is the most common whale in Faroese waters (Bloch, 1996) it may be considered as the major source of *Anisakis*-eggs, delivered with the whale faeces and originating from sexully mature females within the stomach of the pilot whales (Raga and Balbuena, 1993). A measure of the number of visiting pilot whales is the number of whale killed in the traditional Faroese whale hunt called »grindadráp«, taking only the schools coming close to the coast. According to unpublished data from Bloch (1997) the number of pilot whales landed in the years from the 1990 to 1996 were close to the long-term total mean of 844 per year (Bloch, 1996) except the year 1995:

Year	*No. of pilot whales landed*
1990	917
1991	722
1992	1572
1993	804
1994	1201
1995	228
1996	1524

Judging from this data series it is unlikely that the supply of *Anisakis*-eggs from pilot whales has been declining in the period of the observed decline (and rise) in the *Anisakis*-infection of saithe.

Aspholm *et al.* (1995) suggested that only a small population of seals was required to maintain a high level of infection with *Pseudoterranova decipiens* in cod, *Gadus morhua*. Apparently this is also the case for the *Anisakis*-pilot whale system. Whether or not many or few whales are visiting presumably they are able to shed enough number of eggs to enter the food web through the euphausiaceans to saithe.

Encapsulated nematodes in fishes generally are considered to live for months or years. In this study of 1-2 year old saithe calcified nematode larvae, presumably killed by the immune response of the host, were rarely observed. The encapsulated nematodes found of *A. simplex*, *P. decipiens* and *Contracaecum* sp.) are thus following a stable, secure route towards reaching their final hosts through the infection of saithe, while the non-encapsulated larvae and adults of *Hysterothylacium aduncum*, (a low percentage of larvae were found encapsulated), *Cucculanus* sp. and *Echinorhyn-*

chus gadi give an impression which is much more varied (Fig.6c,d). These parasites were mainly found in the gut, which is a very unstable environment.

Polyanski (1966) in an extensive parasitological research of fishes from the Barents Sea reported 45.5 % of 1-year old saithe and 75.0 % of 2-year old saithe infected in/on the liver with *Anisakis* sp. The same figures for *Contracaecum* sp. were 3-4 times higher than in the present study, but he did not find *P. decipiens*. Polyanski (1966) examined seasonal variations of infection for cod and haddock, but not for the saithe.

Scott (1985) investigated the helmints of the alimentary tract of Canadian pollock (=saithe, *Pollachius virens* L.). His results do not clearly distinguish between the years examined, but the pooled prevalence of infection of *A. simplex* show a rapid increase in the first 3 length groups (approximately corresponding to ages 0-3 years) up to 100 %; then a fall for the following length groups (presumably 4-6 years old) down to 77 %. Neither *H. aduncum* nor *P. decipiens* were found, but McClelland *et al.* (1990) reported the prevalence of infection of Canadian pollock, length groups 31-71+ cm, to be 6-11 % for *P. decipiens* , and 89-100 % for *A. simplex*. Comparing these figures with the present study it seems that the main trends in the infections are similar with age, but there are some distinct dissimilarities in the parasite species found on each side of the Atlantic Ocean. More similarity is found when comparing with Icelandic saithe seems to be more similar; Hauksson (1992) reported prevalences of infection to be 72 % with *A. simplex*, 44 % with *Contracaecum* and 44 % with *Hysterothylacium*, but no *P. decipiens* in 26-39 cm saithe (most likely 1-2 years old). The relatively high prevalence of infection up to 60 % with *P. decipiens* in young Faroese saithe (Fig.4 b) thus seems exceptional.

Wootten (1978) investigated five species of offshore, small gadoid fishes (cod, whiting, haddock, Norway pout and poor cod, all primarily 0-2 years old) and found three types of nematodes: *Hysterothylacium aduncum* (then called *Thynnascaris aduncum*), *Anisakis* sp. and *Contracaecum* sp. He did not find *Pseudoterranova decipiens* (then *Phocanema decipiens*), because this nematode requires seals as final host and seals are more commonly found in inshore waters. Interestingly Wootten (1978) recorded more *Hysterothylacium* than *Anisakis* in all five fish species, both in mean intensity and prevalence of infection. He, too found a peak for *Hysterothylacium* in October (compared to June/July). However, Wootten (1978) did not collect nematodes from the gut lumen and the present study is not taking into account stomach nematodes, so the figures are not completely comparable. Also it is difficult to recover all *Anisakis* of the musculature in a dissection, while a digestion technique is more reliable in discovering the hidden nematodes and other multicellular parasites.

An examination of the diet of the saithe would give valuable information in the search to obtain a more detailed knowledge in the correlation of the parasite infection to the composition of the food. Such work could throw more light on the other intrigu-

ing questions of the complex life cycle of *Anisakis simplex*.

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