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STOMACH NEMATODES OF HARBOUR SEAL (PHOCA VITULINA) FROM THE
GERMAN AND DANISH WADDEN SEA

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ABSTRACT

Prevalence of infection with stomach nematodes of 184 harbour seals (Phoca vitulina) of the Schleswig-Holstein part of the German Wadden Sea and of 90 harbour seals of the Danish west coast were investigated. Most of the animals were collected from March to October 1988 during the seal epidemic in the North Sea.

Because seals from the two areas obviously belong to the same population, both samples were pooled together.

The total prevalence of infection was clearly correlated with the age of seals. It increased from 28% in animals younger than 1 year up to 95% in animals older than 2 years.

In hosts older than 2 years, 95% of all seals investigated were infected by Pseudoterranova decipiens, 28% by Contracaecum osculatum, and 13% by Anisakis simplex.

About 95% of all 4953 nematodes found were P. decipiens; about 3% were C. osculatum and about 1% were Anisakis simplex. There was no significant difference in the relative abundance of the three nematode species between the two sexes of seals.

Also the mean number of nematodes per infected seal increased with the age of the hosts. In specimens younger than one year the average number of nematodes per infected seal was 9, in specimens older than 2 years the value was 36. The individual range was 1 to 159 nematodes per seal. An exceptional high number of 719 nematodes found in one seal from Schleswig-Holstein had not been included in the calculation of the mean number.

INTRODUCTION

Five nematode species (Pseudoterranova decipiens, Contracaecum osculatum, Anisakis simplex, Phocascaris phocae and Phocascaris cystophorae) are known from the digestive tract of harbour seals (Phoca vitulina) in the Netherlands (van den BROEK 1963; van THIEL 1966), Great Britain (YOUNG 1972), Iceland (PALSSON 1987), Norway (BENJAMINSEN et al. 1978; BJØRGE 1984) and Canada (SCOTT 1953; SCOTT and FISHER 1958; SCOTT and BLACK 1960; McCLELLAND 1980a-c). However not in every region each of these five species is present.

Regarding the North Sea, the interest in the knowledge of these parasites has grown since the "fish worm crises" in Germany in 1987 and the seal epidemic in 1988.

The eggs of the nematodes are shed with the faeces of the marine mammal host (final hosts: seals or cetaceans) (Fig. 1). The adhesive eggs of P. decipiens sink rapidly and stick to the bottom (BRATTEY 1989), while the eggs of A. simplex will float in sea water (NAGASAWA 1989). Within the eggs the first larva develops and moults to the second stage larva. The free living second stage larva is eaten by marine invertebrates (first intermediate host: mainly crustaceans: decapods and/or euphausiids). The third larval stages of these nematode species are infecting the viscera and flesh of various kinds of fish and therefore reduce their commercial value (second intermediate or transport host). They can be harmful to human health when swallowed alive by consuming raw or unsufficiently cooked fish (see WILLIAMS and JONES 1976). The life cycle of the nematodes is closed when the fish is eaten by a marine mammal, where the parasite moults to 4th stage larva and adults.

Referring to ANONYMOUS (1989) the stock size of harbour seals in the whole Wadden Sea area increased from about 3600 animals in 1974 to about 10500 in August 1988. In Schleswig-Holstein it increased during the same time from about 1500 to about 4000 individuals and in the the Danish part of the Wadden Sea the number increased from about 600 in 1979 to about 1400 in 1987 (1070 in 1988). The population of harbour seals began to break down since sommer 1988 because of an epidemic which was mainly caused by a morbilli virus (OSTERHAUS and VEDDER 1988a; OSTERHAUS et al. 1988b).

Because the harbour seals of the German and Danish part of the Wadden Sea belong to the same population and because there is no natural border between the two sampling areas, the specimens of both areas were pooled together in the following calculations.

MATERIAL AND METHODS

The total prevalence of infection with stomach nematodes was determined by investigation of 184 harbour seals from the Schleswig-Holstein part of the German Wadden Sea and of 90 harbour seals from the Danish part of the Wadden Sea (Fig. 2). Other calculations, such as intensity of infection, relative abundance of nematodes etc., are based on a lower number of seals (as indicated in the figures), depending on the availability of seal data (age, sex, date of finding).

In 90 of the 184 seals from German waters the oesophagus could be examined in addition to the stomach, and in 81 seals also the intestine could be examined. Seals from Danish coast were examined without oesophagus. In 84 of the 90 Danish seals also the intestine was examined.

The Danish material was collected from May to October 1988 during the seal epidemic. The German specimens were collected from July to October in the same year. Additional 12 stomachs were examined from seals found dead from March to April 1989. For calculations of monthly changes, all seals were pooled together in one annual cycle. The highest numbers of specimens investigated were found from July to September (Fig. 3). From all 274 harbour seals examined 14 were euthanized (mostly pups), the rest was found dead on sands or beaches.

Age of seals was estimated by the help of length, weight and date of finding (Ag = Age group):

- Ag1 : seals younger than 1 year
- Ag2 : seals between 1 and 2 years old
- Ag3 : seals older than 2 years

The age and sex composition of the seals examined is shown in Figure 4.

Most stomachs were investigated after freeze storage, only a few samples could be studied fresh. The nematodes were transferred to physiological NaCl-solution and were examined on the same day by using a binocular microscope. So there was no need for making the worms transparent by the help of clearing chemicals.

For identification of the nematode species, differences in the morphology of intestine, lips and spicules were used as main characteristics (DELYAMURE 1955; MYERS 1960).

RESULTS

Distribution of Nematodes within the Digestive Tract

In 90 seals the whole digestive tract was searched for parasites. Nematodes were found in 69 seals (77%) in the stomach, in 19 seals (21%) in the oesophagus and in 2 seals in the intestine (2%; within the first meter behind the stomach exit). The abundance of nematodes in the different parts of the digestive tract was not counted, but, as an estimation it can be said that the vast majority (more than 90%) of the worms was always present in the stomach and less than 10% were found in the oesophagus. In hosts with no nematodes in the stomach, also no nematodes were present in any other part of the digestive tract. The presence of nematodes in the oesophagus was correlated with the age of seals. In 16 out of 43 infected seals (37%) older than 2 years nematodes were found in that part of the digestive tract. In Ag2 only 3 out of 17 infected hosts (18%) had nematodes in the oesophagus and in Ag1 no nematodes were present in that organ (9 infected seals).

The nematodes usually were found free in the stomach. Only in a few cases nematodes were attached to the stomach wall. These nematodes mostly belonged to the genus Contracaecum. In two cases no worms but the caps of the molting larvae (as described by McCLELLAND and RONALD 1974) could be detected in the stomach mucosa.

Total Prevalence of Infection

The total prevalence of infection was clearly correlated with the age of seals. Newborn and suckling pups had no stomach nematodes. 28.1% (25/89) of the seals were infected by nematodes in Ag1, 79.7% (63/79) in Ag2 and 94.8% (92/97) in Ag3 (Fig. 5). There was no clear difference in the total prevalence of infection between the two sexes of seals (Fig. 5).

Concerning the present material no clear seasonal changes in the infection rate could be detected with the exception of June and July (Fig. 6). In June none of the 5 seals of Ag1 was infected with nematodes. The two infected seals of Ag1 in July carried only larval stages of nematodes. The first adult stages of nematodes in the stomach of specimens of Ag1 were detected in August.

Prevalence of Infection for different Nematode Species

Three morphologically distinguishable species of nematodes were found: Pseudoterranova decipiens, Contracaecum osculatum and Anisakis simplex. For all these species there was a clear increase in infection prevalence with increasing age of seals (Fig. 7). In Ag1, P. decipiens was found in 29.4%, in Ag2 it was present in 78.8% and in Ag3, 94.3% of the seals were infected with this nematode species. The values of infection prevalence with C. osculatum were 8.8% in Ag1, 19.7% in Ag2 and 27.6% in Ag3. The infection prevalence with Anisakis increased from 2.9% in Ag1 to 12.6% in Ag3.

Intensity of Infection

The number of nematodes found in single seal stomachs ranged between 1 and 159 in German samples and 1-126 in Danish samples. An exceptionally high value of 719 nematodes (mostly L3 and L4 larvae) was found in the stomach of an adult male seal (sampled in Schleswig-Holstein in August 1988). This animal was excluded from the following calculations of the infection intensity.

The mean intensity of nematodes per infected seal increased with the age of the hosts (Fig. 8, Table 1). The mean value increased from 9 to 36 (Ag1 to Ag3, respectively). In Ag1 the majority of the seals was not infected or only had a low number (1-10) of nematodes in their stomachs. In Ag2 most of the seals were infected by 1-10 nematodes and in Ag3 the majority of seals was infected by 1-20 nematodes per stomach (Fig. 12a-c).

The mean abundance of nematodes in all seals (= mean number of all seals investigated) is also given in Table 1.

There was no clear correlation between the mean intensity of nematodes and the sex of seals.

Relative Abundance of different Nematode Species

The most common nematode species was Pseudoterranova decipiens. 93.4% of all 4953 nematodes found belonged to that species, 2.7% belonged to Contracaecum osculatum and only 0.7% to Anisakis simplex. There were only weak differences concerning the different age groups of seals and the two sampling areas (Fig. 9, Table 2). In young seals the relative abundance of Contracaecum was slightly higher (5.6%) than in old ones (2.1%). There were no significant differences in the composition of the different nematode species between the two sexes of seals.

Regarding monthly changes in the composition of the nematode fauna there was a peak for C. osculatum in June/July (18.3/8.5%; Fig. 10, Table 3). In the other months the relative abundance of Contracaecum was lower than 5.3%. However, in June a total number of 57 nematodes only was found in the 15 seals investigated).

Relative Abundance of different stages and sexes of P. decipiens

Compiling all age groups of seals, the ratio between male and female adult nematodes of Pseudoterranova was 1:1.02. Concerning the different age classes it was 1:1.44 in Ag1, 1:1.11 in Ag2 and 0.97:1 in Ag3 (Table 4).

Figure 11 shows the relative abundance of larval and adult Pseudoterranova per month. There is a relatively low percentage of larval Pseudoterranova in April. The value of October has to be neglected because of the low sample size (28 Pseudoterranova in 4 seals). Concerning C. osculatum and A. simplex, the number of specimens found was too low for calculations of monthly changes.

DISCUSSION

Most of the seals investigated for this report came from an epidemic, the main cause of which is supposed to be a morbilli virus (OSTERHAUS and VEDDER 1988a, OSTERHAUS et al. 1988b). As seal hunting in the Wadden Sea is rigorously restricted, this was the only opportunity to obtain large material for parasitological investigations. Moreover it has to be considered that the parasitic fauna of dying seals does not necessarily reflect the parasitological conditions of a "normal" (healthy) seal population.

Because of the absence of other seal species in the sampling area (grey seals are only represented by less than 80 individuals, WEIDEL 1988) and as cetaceans are supposed to be transmitters mainly for Anisakis (YOUNG, 1972), the harbour seal has to be considered as the main final host for P. decipiens in the Wadden Sea. This nematode species represents more than 90% of the total number of worms found. SCOTT and FISHER (1958) also found such a high relative abundance of P. decipiens in harbour seals from the southern Canadian Atlantic coast. The low abundances of C. osculatum and A. simplex indicate that harbour seal does not play an important role in transferring these nematode species to invertebrates and fish in that region. YOUNG (1972) also found only very few Anisakis larvae (7%) and no adult specimens in harbour seals from British waters. The relative abundance of Contracaecum found by this author, however, was 45% in adult nematodes and 71% in larval nematodes. PALSSON (1987) did not find any Anisakis in harbour seals from Iceland. In contrast to that finding, the relative abundance of A. simplex in Norwegian waters was 47%, and 11% for C. osculatum (BJØRGE 1984). These data indicate clearly that the composition of the nematode fauna depends on the area and/or the availability of different food items in different regions.

The most important intermediate or transport hosts, transferring P. decipiens to seals in the German Wadden Sea seem to be smelt (Osmerus eperlanus) and sea scorpion (Myoxocephalus scorpius) (MÖLLER and KLATT, 1988). The relatively low ratio of larval to adult Pseudoterranova in April may be due to the spawning season of smelt in March/April. Adult smelt then migrate from the Wadden Sea area into estuaries and freshwater regions of the North Sea tributary rivers and are less available for predators like the harbour seal.

No fish host is known for C. osculatum in the Wadden Sea. FAGERHOLM (1988) described the third stage larvae from cod (Gadus morhua) from the northern Baltic Sea and PALSSON (1987) found numerous Contracaecum/Phocascaris larvae in capelin (Mallotus villosus) from Icelandic waters. Food analyses of the very few Wadden Sea seals which had food contents in their stomachs showed that in 10 cases were cod had been eaten, only in two seals Contracaecum was found. Capelin is absent in the sampling area.

The total prevalence of infection was clearly correlated with the age of the hosts. In seals younger than one year it depends mostly on the portion of pups in the samples. The pups get first infected with stomach nematodes after they start feeding on fish and perhaps invertebrates. This is the case in July when the pups get weaned about 4-6 weeks after birth (KING, 1983). Therefore, in July only larval stages of nematodes could be found in the stomachs of seals younger than 1 year. For Pseudoterranova, the time to develop to the adult stage is about 5-15 days and to reach maturity it lasts 15-25 days (McCLELLAND 1980a, b; TOWNSLEY et al. 1963). This fits with the first finding of adult stages of P. decipiens in seals of that age group in August. The lower rate of infection in younger seals depends on the feeding behaviour. Younger seals mostly feed on small

fishes such as gobies (Pomatoschistus sp.) and crustaceans (Crangon sp.) and relatively seldom on smelt (BEHREND 1985, SIEVERS 1985). In older seals the portion of highly infected fish species (smelt) is much higher. Therefore, the infection rate of these seals should be expected to be 100%. However, such an infection rate was only reached by the Danish seals older than 2 years. There are different explanations possible: It had been mentioned in other reports that seals may vomit (McCLELLAND 1980b) or that the nematodes may leave the host after its death through the mouth, nostrils or rectum (MYERS 1960). The finding of nematodes in the oesophagus and, in two cases, in the intestine confirms the described behaviour. Another explanation could be the different feeding behaviour and, therefore, feeding on non-infected fish in some individuals. Also a kind of resistance as described for experimental infections by McCLELLAND (1980b) may be an explanation.

The mean intensity of nematodes was also increasing with the age of seals. It ranged from 3.3 in Ag1 (DK) to 39.1 in Ag3 (SH) with a range of 1-10 and 1-159, respectively. Comparing investigations in stomach nematodes of fresh harbour seals from other regions, the mean intensity was 17.7 (range: 5-34) off East Anglia (Great Britain) and 87 (range: 21-182) off the Western Islands of Scotland (after YOUNG, 1972). In Norwegian coastal waters the mean intensity was 60 (BJØRGE, 1984). In Canadian waters SCOTT and FISHER (1958) found the mean value ranging from 20 to 225 depending on the sampling area. The intensity of nematodes, however, never reaches the high level as described for grey seals (Halichoerus grypus, YOUNG 1972; McCLELLAND 1980; BJØRGE 1984).

As mentioned before, stomach nematodes found in the oesophagus of some seals may indicate that the seals had vomited before dying or some nematodes may have left the host after death. Furthermore, it seems to be possible that ill seals may have fasted for an unknown period because they were not able to dive due to emphysema or weakness (BREUER et al. 1988). Only 7% of the investigated seals had food contents or otoliths in their stomach. Therefore, the results in the present report have to be considered as a minimum nematode burden.

The presence or absence of the oesophagus seems to play a less important role in quantifications of stomach nematodes of hosts found dead. It doesn't play a role in determination of the infection prevalence. The examination of the intestine is negligible in this point of view. Also YOUNG (1972) did not find worms in the oesophagus or in the intestine of 19 fresh seals. However, as findings of nematodes in the intestine of other seal species and/or in other regions showed, these parasites can be present in high numbers in that part of the digestive tract (BERLAND, 1963; DELYAMURE and TRESHCHEV, 1966).

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ABBREVIATIONS

A	Anisakis
Ag	Age group
C	Contracaecum
DK	Denmark
F	Female
M	Male
SH	Schleswig-Holstein, F.R.Germany
P	Pseudoterranova

DEFINITIONS

Intensity of Infection: Number of nematodes found in a seal stomach.

Mean intensity: Mean number of parasites per infected seals.

Mean abundance: Mean number of parasites per infected and not infected seals.

Prevalence of Infection: Percentage of seals infected with nematodes.

Range: Minimum and maximum number of nematodes found

Relative abundance: Percentage of different nematode species related to all nematodes found.

TABLES AND FIGURES.

Table 1. Mean intensity and mean abundance of nematodes per seal.

Area	Age group	Range	Mean number	Std. dev.	No. of seals	Range	Mean abundance	Std. dev.	No. of seals
SH	Ag1	1- 62	12.1	16.2	15	0- 62	3.8	10.6	48
	Ag2	1- 94	19.1	23.7	27	0- 94	15.1	22.5	34
	Ag3	1-159	39.1	35.6	84	0-159	38.3	35.7	85
DK	Ag1	1- 10	3.3	3.5	6	0- 10	0.9	2.3	23
	Ag2	1-105	18.0	21.7	28	0-105	16.3	21.3	31
	Ag3	1-126	30.6	29.3	32	0-126	30.6	29.3	32
both	Ag1	1- 62	9.6	14.4	21	0- 62	2.8	8.9	71
	Ag2	1-105	18.5	22.7	55	0-105	15.7	21.9	65
	Ag3	1-159	35.8	33.6	116	0-159	35.4	33.6	117

Table 2. Relative Abundance of the different Nematode Species.
(n = Total Number of Nematodes per Age Group)

Age group	G e r m a n y				D e n m a r k			
	Pseudo	Contra	Anisakis	n	Pseudo	Contra	Anisakis	n
Ag1	93.8%	5.7%	0.6%	181	90.0%	5.0%	5.0%	20
Ag2	94.8%	4.3%	1.0%	515	94.1%	4.6%	1.4%	505
Ag3	97.8%	1.5%	0.6%	2711	96.0%	3.7%	0.3%	913
All Ag	92.7%	2.1%	0.7%	3481	95.3%	3.9%	0.8%	1472

Age group	B o t h A r e a s			
	Pseudo	Contra	Anisakis	n
Ag1	93.4%	5.6%	1.0%	201
Ag2	94.4%	4.4%	1.2%	1020
Ag3	97.4%	2.1%	0.6%	3624
All Ag	96.6%	2.7%	0.7%	4953

Table 3. Relative abundance of different nematode species per month. (All nematodes found per month were set 100%; German and Danish specimens pooled together).

Month	Pseudo.	Contra.	Anisakis
Jan	—	—	—
Feb	—	—	—
Mar	92.0%	2.0%	6.0%
Apr	94.7%	5.3%	0.0%
May	96.6%	0.9%	2.6%
Jun	80.3%	18.3%	1.4%
Jul	89.3%	8.5%	1.7%
Aug	97.7%	1.6%	0.5%
Sep	97.5%	1.4%	0.5%
Okt	96.6%	3.4%	0.0%
Nov	—	—	—
Dec	—	—	—

Table 4. Relative abundance of adult P. decipiens (male/female) in the different age groups and sexes of harbour seals. (German and Danish specimens pooled together).

Sex of seals	Age groups	P. decipiens	
		Males	Females
both sexes	AG 1	41.0%	59.0%
	AG 2	47.4%	52.6%
	AG 3	50.7%	49.3%
Male seals	Ag1	38.6%	61.4%
	Ag2	50.0%	50.0%
	Ag3	50.6%	49.4%
Female seals	Ag1	42.9%	57.1%
	Ag2	46.5%	53.5%
	Ag3	50.8%	49.2%
both sexes	All Ages	49.5%	50.5%

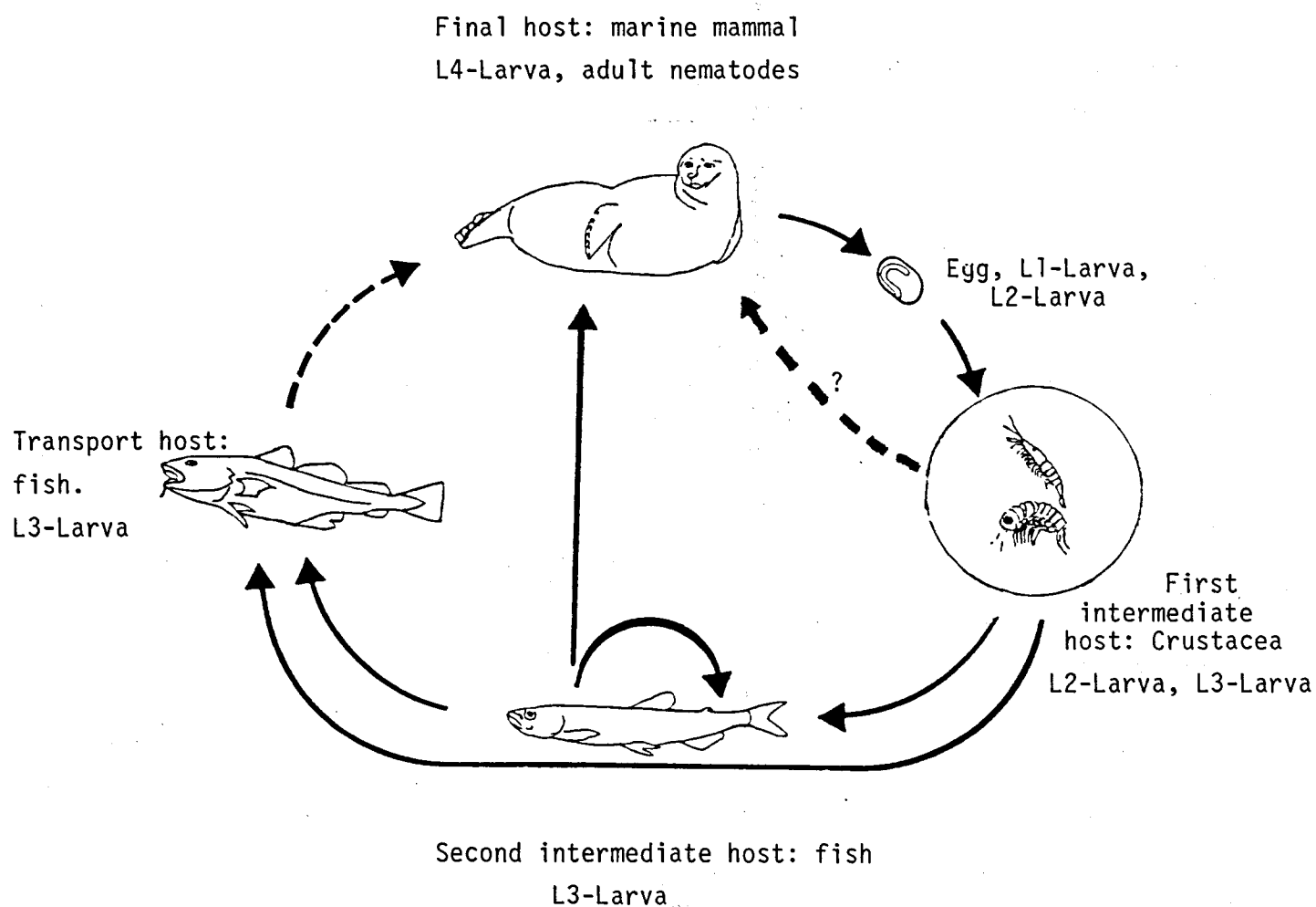


Figure 1. Principle life cycle of Anisakine nematodes (after KLATT 1985, redrawn).
The eggs of Pseudoterranova decipiens sink to the bottom, become adhesive, and stick to the substrate. The eggs of Anisakis simplex float in sea water.

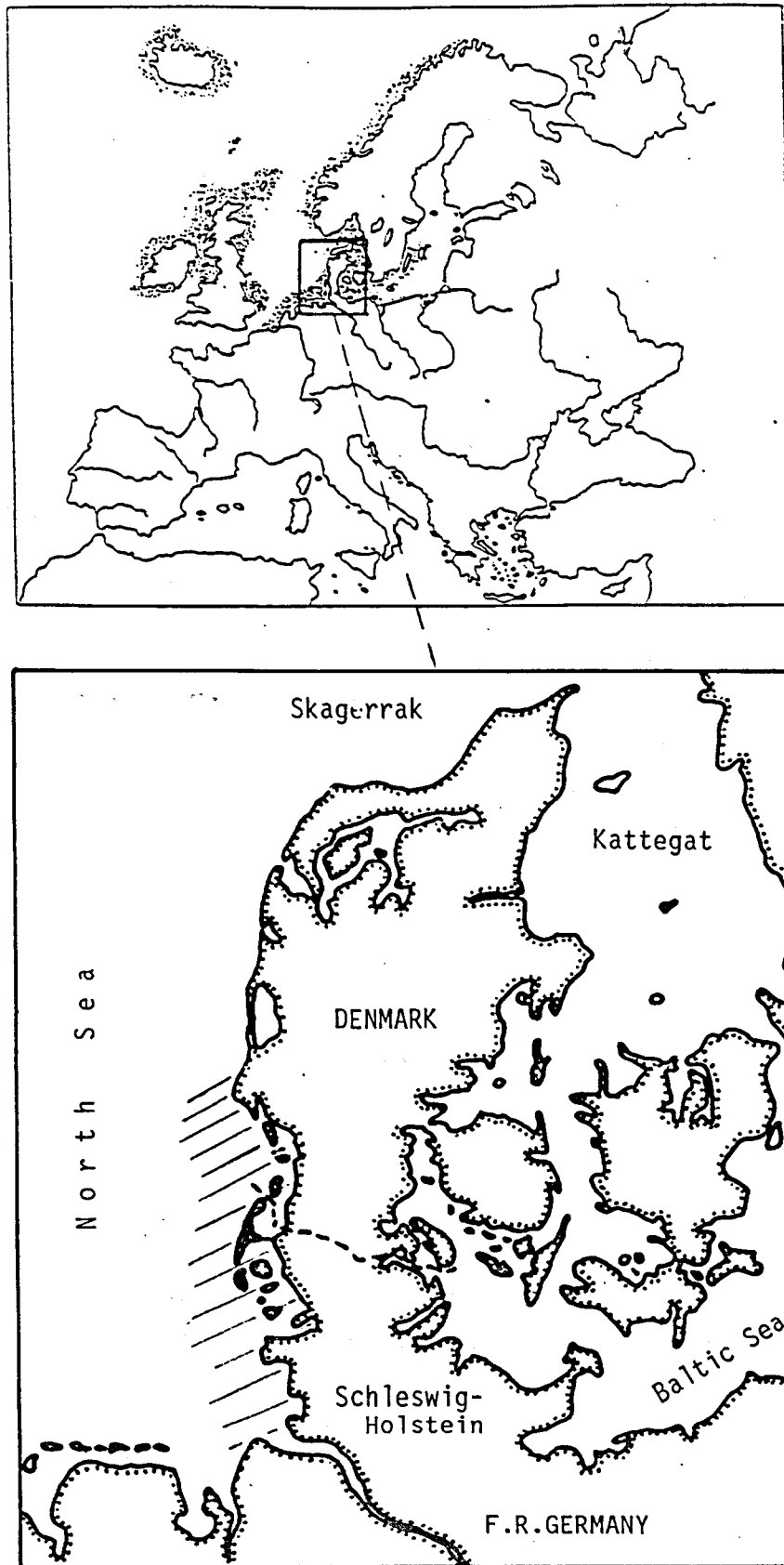


Figure 2. Distribution of harbour seal (*Phoca vitulina*) in Europe (upper Figure, after HEIDEMANN, cit. in ANON 1989) and sampling area in the German and Danish part of the Wadden Sea (////).

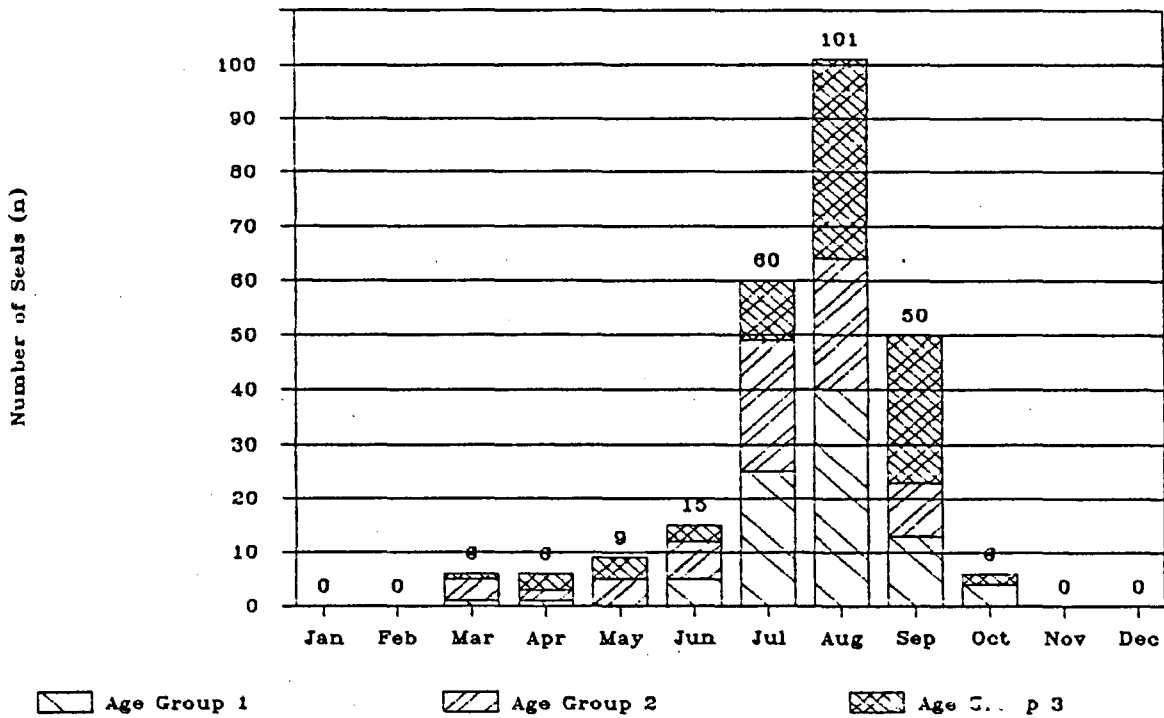


Figure 3. Number of harbour seals investigated per month (German and Danish specimens pooled together, n=253).

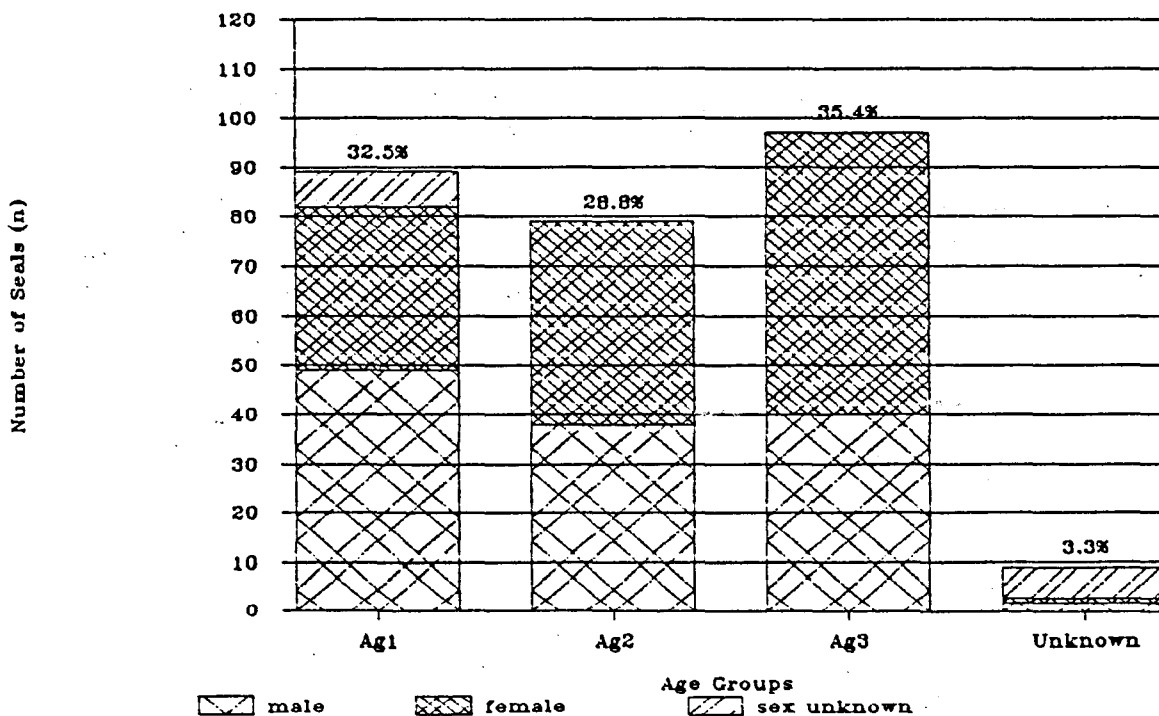


Figure 4. Age and sex composition of harbour seals investigated (German and Danish specimens pooled together, n=274).

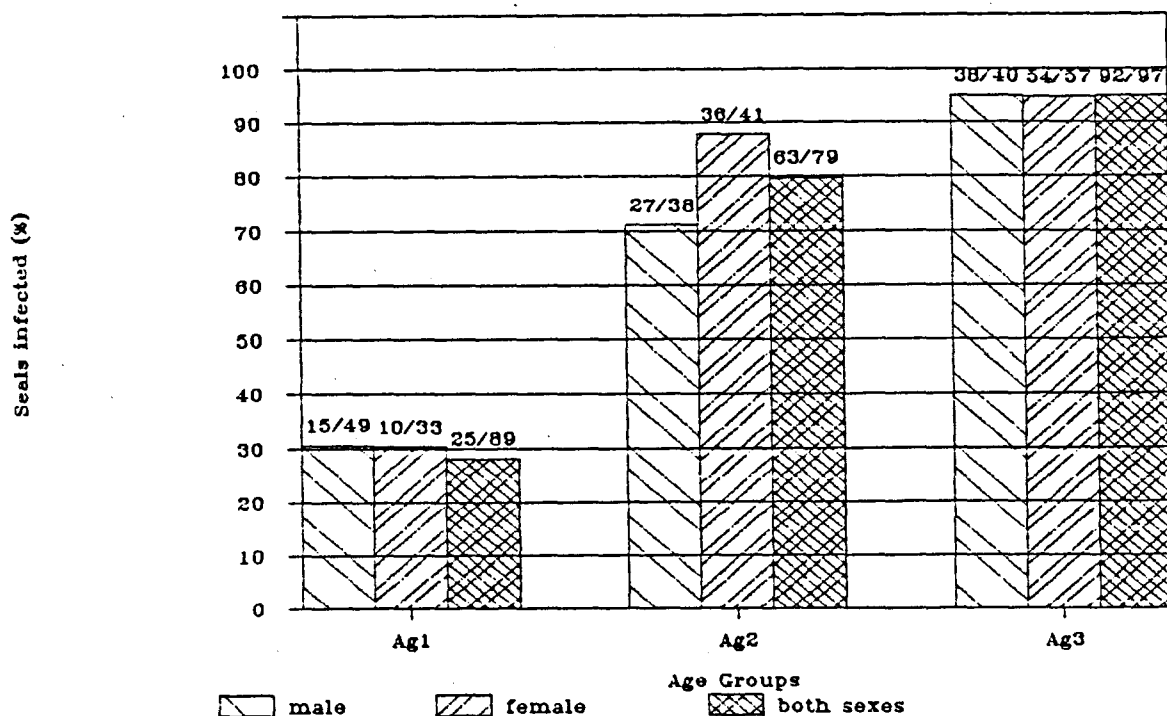


Figure 5. Total prevalence of infection of the different age groups and two sexes of harbour seals. (German and Danish seals pooled together, n=265)

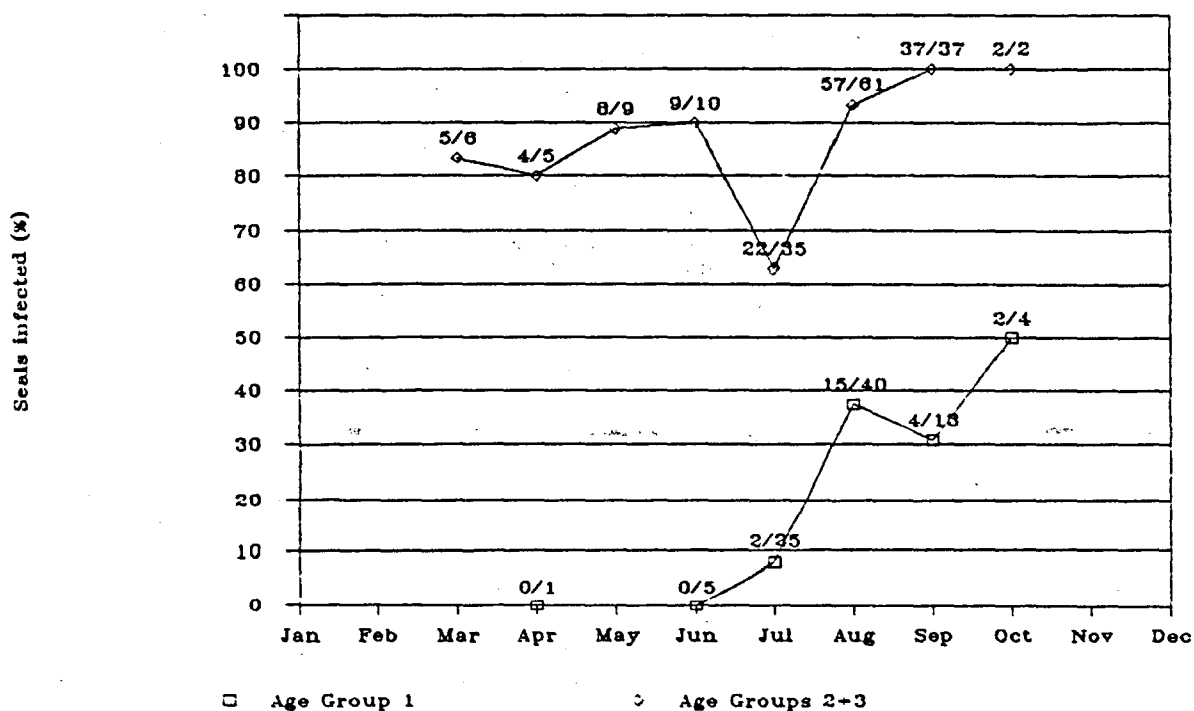


Figure 6. Monthly changes in total infection prevalence (German and Danish specimens pooled together, n=253)

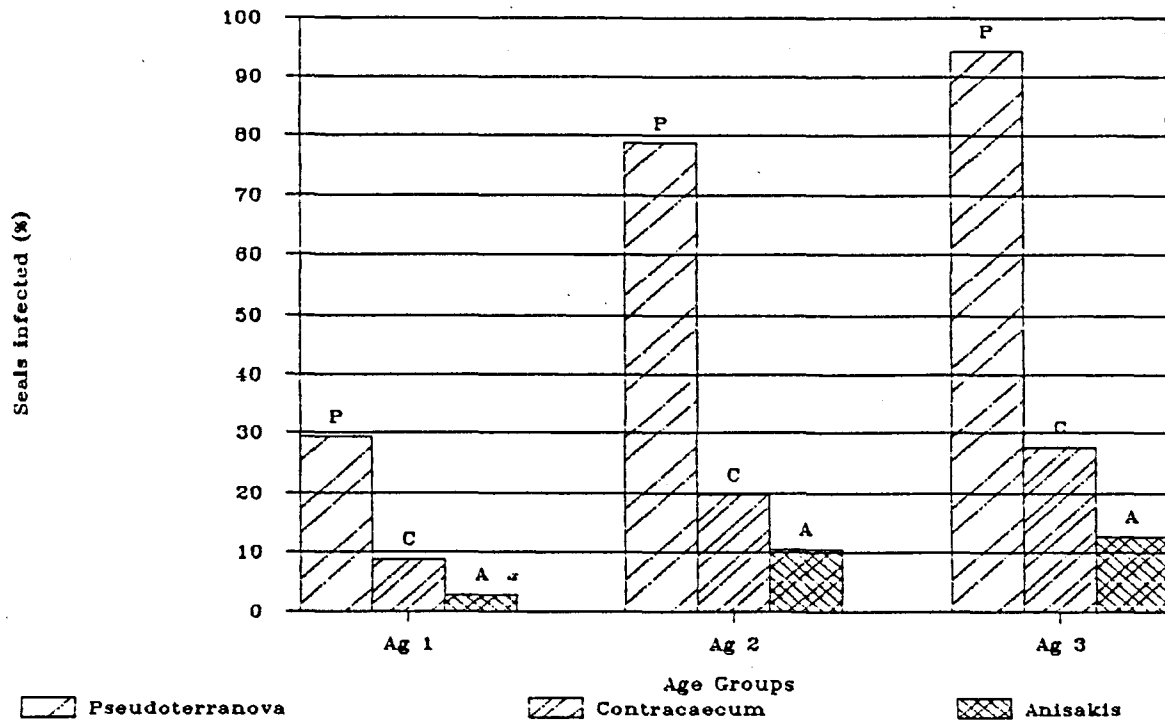


Figure 7. Infection prevalence with different nematode species (German and Danish seals pooled together, n=233).

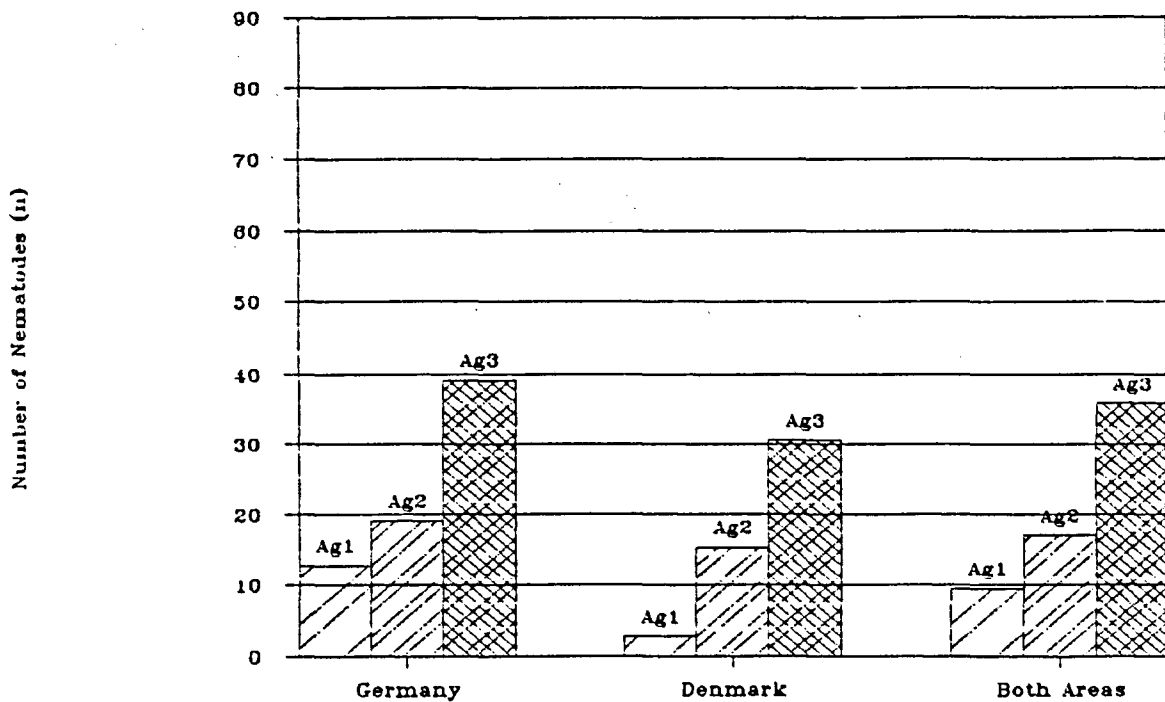


Figure 8. Mean number of nematodes per infected seal in the different age groups of harbour seals. (German seals investigated: n= 73; Danish seals: n= 64). An exceptional finding of 719 nematodes in one seal stomach in Germany is excluded in this figure.

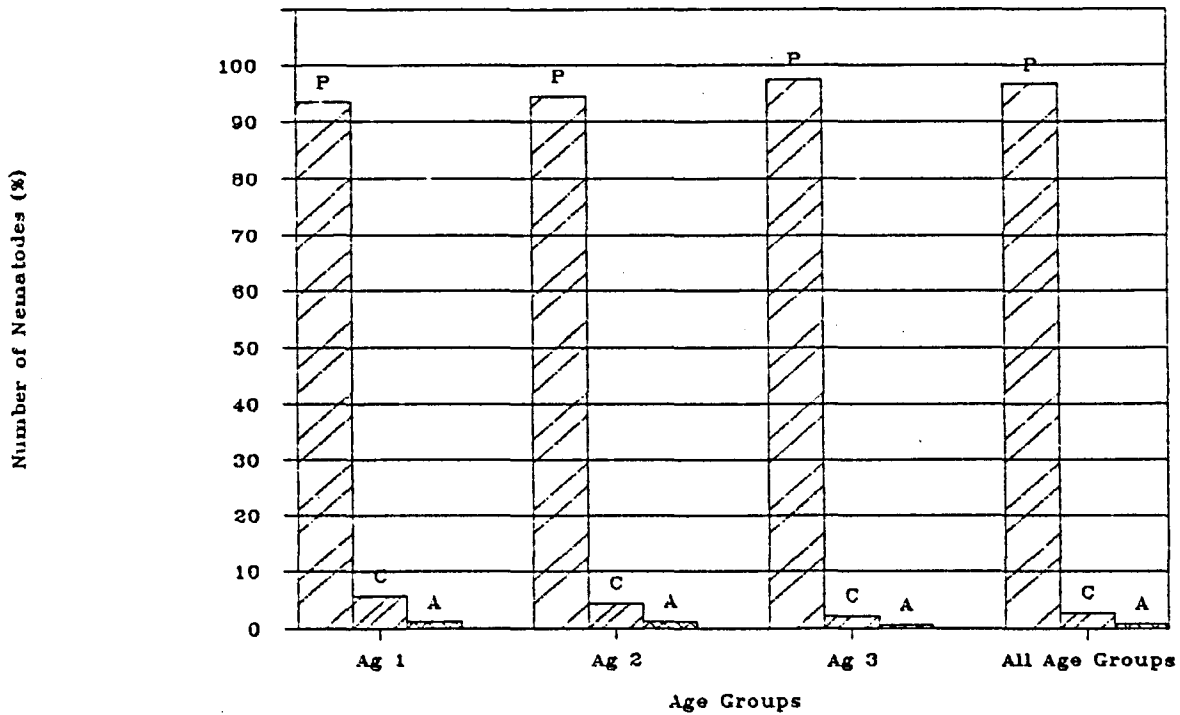


Figure 9. Relative abundance of the different nematode species in the different age groups of seals. (German and Danish specimens pooled together; total number of nematodes found, n=4953).

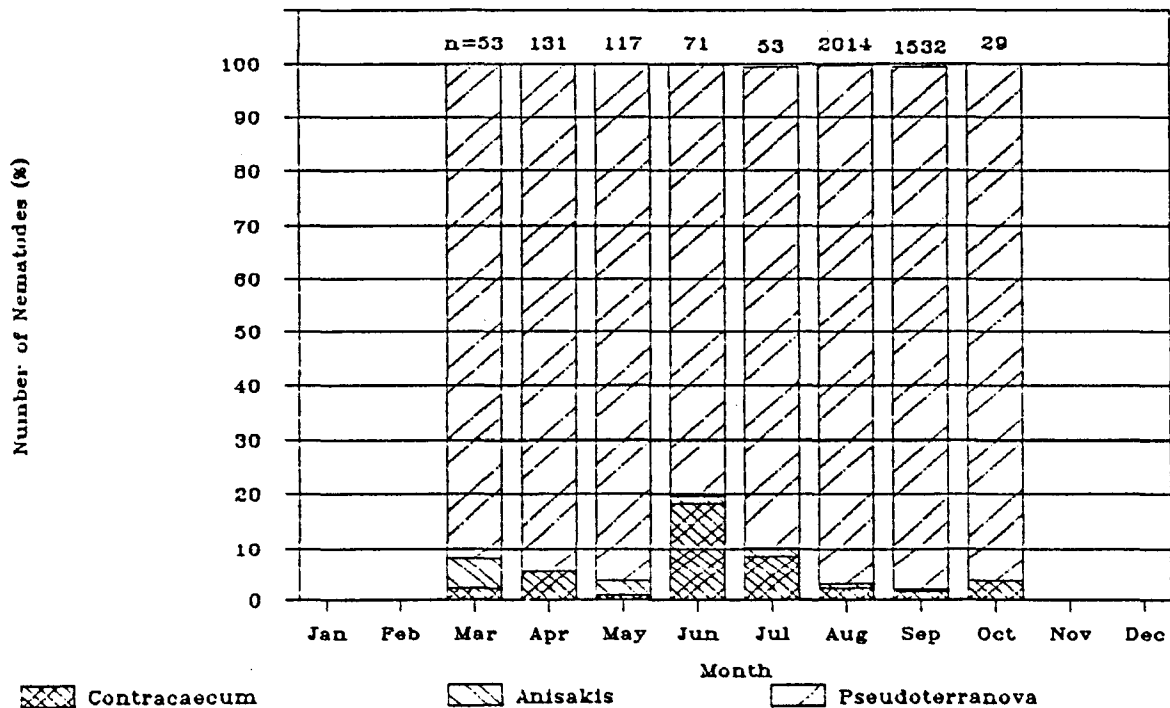


Figure 10. Relative abundance of different nematode species per month. (German and Danish specimens pooled together; n = number of nematodes per month, = 100%).

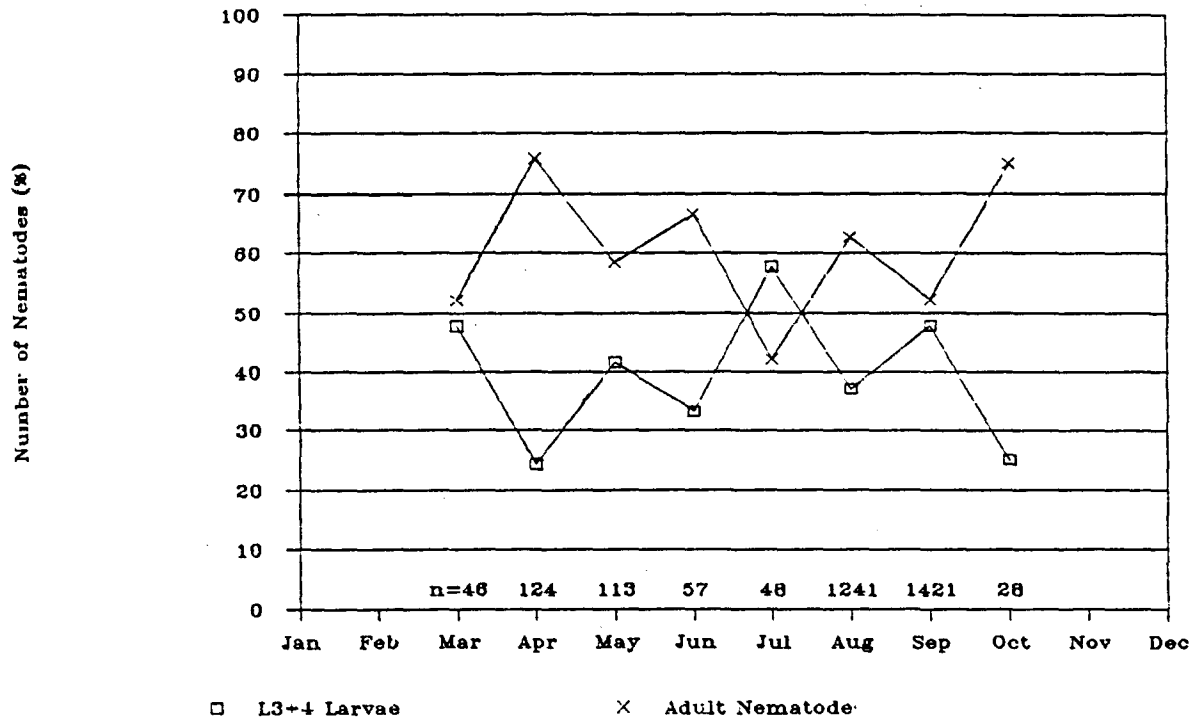


Figure 11. Relative abundance of larval (L3+L4) and adult Pseudoterranova per month (German and Danish specimens pooled together).

Fig. 12a.

Number of Seals infected

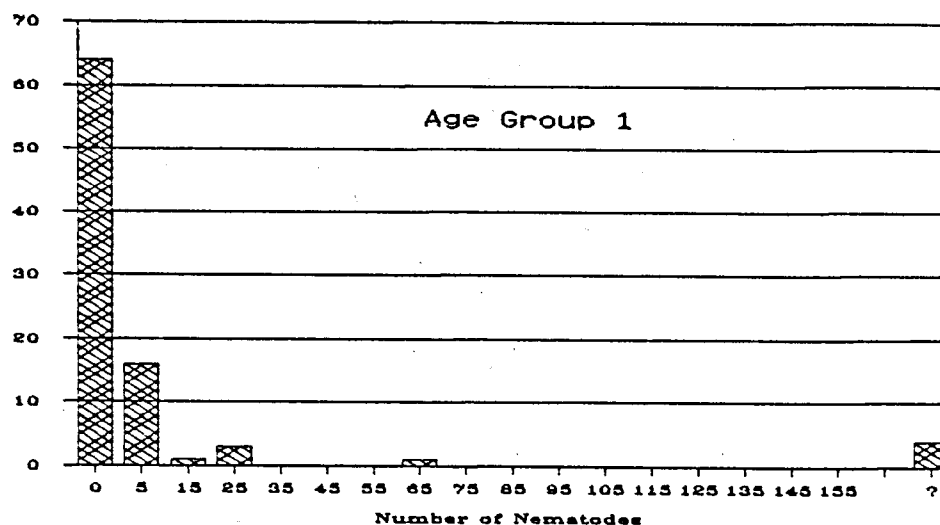


Fig. 12b.

Number of Seals infected

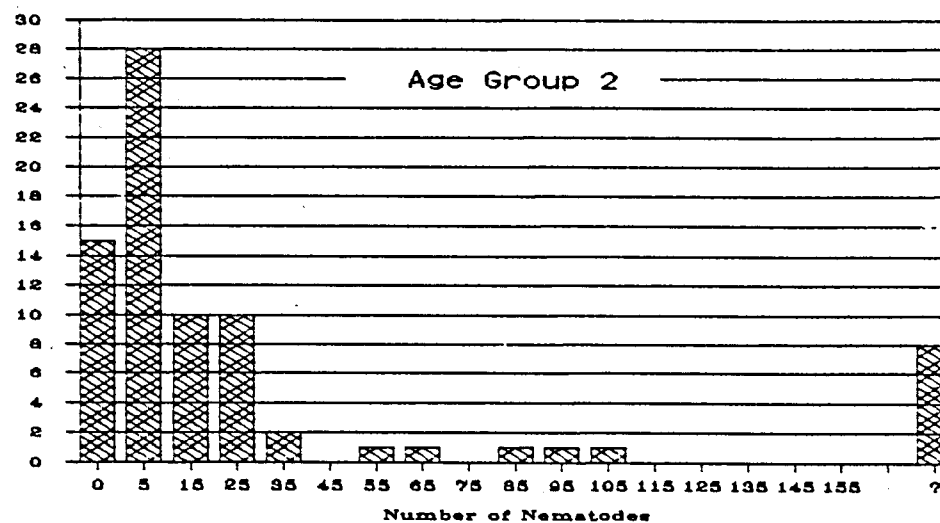


Fig. 12c.

Number of Seals infected

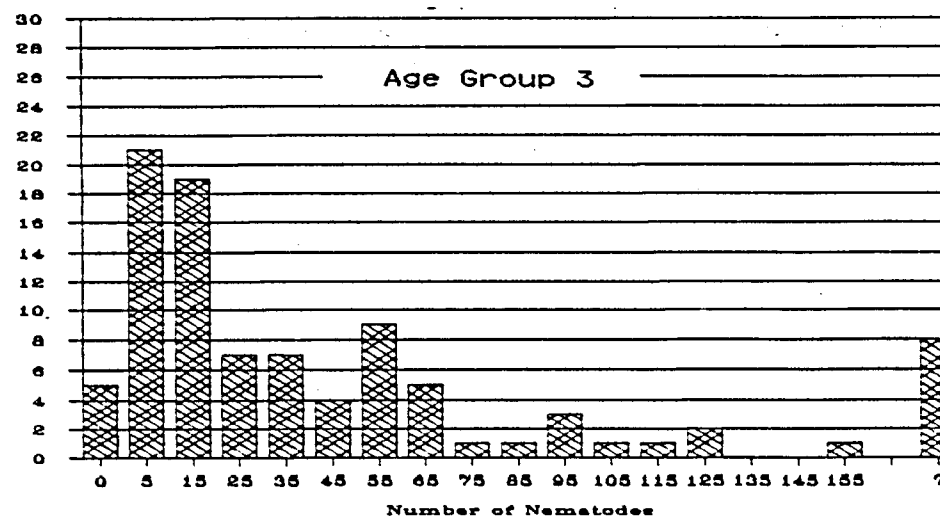


Figure 12 a-c. Frequency distribution of seals of different age groups infected with nematodes (grouped into 10; x-scale indicates $x \pm 5$ nematodes). An exceptional finding of 719 nematodes in one seal stomach in Germany is excluded in Figure 12c.