# Occurrence of Anisakis sp. larvae in the chilean Jack Mackerel, Trachurus murphyi Nichols 1920

# Ocurrencia de larvas Anisakis sp. en el jurel Trachurus murphyi Nichols 1920

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## ABSTRACT

The presence of larval anisakids was investigated in 120 Trachurus murphyi from Talcahuano, off the coast of Chile. Forty five percent of the fish was infected with 208 Anisakis sp. (type I) larvae. Parasite intensities ranged from one to 22 larvae per infected fish. This is the first time the intensity of infections with Anisakis sp. larvae is reported in Trachurus murphyi. Statistical analysis was made with non-parametric tests. Prevalence and intensity of infections increased with length of fish. Sexual maturity of fish seems to play a role in the prevalence and intensity of infections: there were no significant differences between immature fish, but sexually mature females had significantly higher prevalence and intensity of infections occurred on gonadic mesenteries and only three fish (the largest in the sample) were infected in the muscles. The possible influence of sexual maturity on prevalence and on intensity of infections, statistical procedures employed in the analysis of data, and location of parasites within the host body are the main topics discussed.

Keywords: Anisakis, parasite, infection, Trachurus murphyi, Chile.

#### RESUMEN

Se investigó la presencia de larvas de anisákidos en 120 jureles Trachurus murphyi colectados en Talcahuano, Chile. El 45% de los peces estaba infectado, y portaban un total de 208 larvas de Anisakis sp. (Tipo I). Las cargas parasitarias fluctuaron entre 1 y 22 larvas por pez infectado. Este es el primer registro de las intensidades de infección por larvas de Anisakis sp. en el jurel Trachurus murphyi. El análisis estadístico fue realizado mediante pruebas no paramétricas. La prevalencia e intensidad de las infecciones se incrementaba con la talla de los peces. La madurez sexual de los peces parece ser un factor importante en los valores de prevalencia e intensidad de las infecciones: no había diferencias significativas entre los peces inmaduros, sin embargo, las hembras sexualmente maduras tenían prevalencias e intensidades de infección significativamente mayores que los machos (P < 0.05). La mayoría de las infecciones ocurría en los mesenterios gonadales, y sólo tres peces (los más grandes de la muestra), estaban infectados en la musculatura.

Los principales tópicos discutidos son, la posible influencia del estado de madurez sexual del pez sobre la prevalencia e intensidad de las infecciones, los procedimientos estadísticos empleados en el análisis de los datos y la localización de los parásitos en el cuerpo del huésped.

Palabras-clave: Anisakis, parásito, infección, jurel, Chile.

Larval nematodes of marine fishes have been frequently studied in recent years because of the occurrence of clinical human cases associated with Anisakidae larvae acquired through consumption of raw or insufficiently cooked marine fish. Human infections have been reported in North America, Japan, The Netherlands, Scandinavian countries (Williams & Jones 1976, Smith & Wootten 1978) and in Chile (Sapunar et al. 1976, Apt et al. 1980). The damage that these parasites may cause to fish hosts has also been reviewed (Margolis 1970, Cheng 1976).

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Anisakis sp. larvae have been reported from more than 200 species of marine fishes in different oceans of the world. Most of the reports are from clupeids in the North Atlantic, North Pacific and North Sea (Bishop & Margolis 1955, Khalil 1969, Davey 1972, Van Banning & Becker 1978). Salmonid fishes (Novotny & Uzmann 1960, Margolis 1963, Beverley-Burton & Pippy 1978) and gadoid fishes (Grabda 1976, Young 1972, Wootten & Waddell 1977, Carvajal et al. 1979) have also been frequently investigated. A major review on this subject is that by Smith & Wootten (1978). At least 12 marine fishes from the Southeast Pacific Ocean have been reported as hosts for anisakids (George-Nascimento & Carvajal 1980).

Although *Trachurus murphyi* Nichols 1920 is an important marketable fish in Chile and Peru, there are no previous reports on the intensity of infections with *Anisakis* sp. larvae. The present paper deals with both prevalence and intensity of infections in relation to sex, length, gonad development stage of the host, and location of parasites within the host body.

### MATERIAL AND METHODS

One hundred and twenty jack mackerels, *Trachurus murphyi* Nichols, 1920 were collected between April and December 1977. Fishes were obtained from commercial fishing trawlers operating between the vicinity of Dichato (36°33'S; 73°56'W) and Lebu (37°37'S; 73°41'W), off the coast of Talcahuano, Chile.

Standard length was obtained for each specimen. Sex and sexual maturity stage were determined according to Kayser (1973). Then the fish in the sample were divided into two groups: (a) those with undeveloped or resting gonads (immature), and (b) those with gonads in the process of development or spawning (mature).

Parasites found in the coelomic cavity and viscera of fishes (except the liver) were removed and preserved in 10% formalin for later identification. Muscles and liver of fishes were artificially digested following a technique similar to that of Novotny & Uzmann (1960). Larvae obtained by diges-

tion were also preserved in 10% formalin. All larvae collected were cleared in Amman lactophenol for microscopic examination. Parasites were identified up to genus level, according to the criteria of Berland (1961).

Data were analyzed statistically using mainly non parametric methods. The Miller Jackniffe test for analysis of variance was preferred to others because of its ability in reducing the bias of point estimators; in addition, it has been recommended where deviations from normality of the underlying population can be disastrous for the classical F test for equal variances, and where distribution free rank tests for the problems are limited in their applicability (Hollander & Wolfe 1973).

Prevalence of infections was measured as the percentage of fish infected in a sample. The analysis of intensity distributions considered only infected fishes because a cummulative nature of piscian anisakiasis was assumed, as several authors have suggested, (Bishop & Margolis 1955, Novotny & Uzmann 1960, Khalil 1969, Cheng 1976). This assumption implies that uninfected fish have never harboured parasites.

## RESULTS

Fifty four (45%) out of 120 fishes examined were infected with Anisakis sp. larva (I), as designated by Berland (1961). These parasites were mainly located in the coelomic cavity, encapsulated and attached to the mesenteries. Infected fish were larger than non-infected ones (Wilcoxon rank sum test; P < 0.01). Data showed increasing values of prevalence of infection with increasing length of the host (Table 1).

Intensities of infection in 51 out of the 54 infected hosts were analysed; data on three fish (two females and one male) were lost. A total number of 208 Anisakis sp. larvae were collected ( $\bar{x} = 4.08$  larvae/ infected fish; SD = 4.07). Frequency distribution of intensities was skewed and overdispersed. All attempts to fit the data to the negative binomial, truncated negative binomial, positive binomial and logarithmic series were unsuccessful. A positive correlation was found

TABLE 1
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Occurrence of Anisakis sp. larvae in Trachurus murphyi, according to length and sex of the host. Ocurrencia de larvas Anisakis sp. en Trachurus murphyi, según la longitud y sexo del huésped.

	BOTH	SEXES	FEM	ALES	MA	LES
Fish Length (cm)	N <sup>o</sup> of fish examined	N <sup>o</sup> of fish infected	N <sup>0</sup> of fish examined	N <sup>o</sup> of fish infected	N <sup>0</sup> of fish examined	N <sup>0</sup> of fish infected
28.7 - 30.6	1	0	0	_	1	0
30.7 - 32.6	0	-	_	_	_	
32.7 - 34.6	5	0	2	0	3	0
34.7 - 36.6	34	7	26	5	8	2
36.7 - 38.6	36	16	17	10	19	6
38.7 - 40.6	22	13	16	10	6	3
40.7 - 42.6	12	9	6	5	6	4
42.7 - 44.6	2	2	1	1	1	1
44.7 – 46.6	4	3	2	2	2	1
46.7 - 48.6	4	4	3	3	1	1
TOTAL	120	54	73	36	47	18

between intensities of infection and length of infected fish (Spearman correlation coefficient:  $r_s = 0.56$ ; P < 0.01; Fig. 1).

Eighteen males (38%) and 36 females (49%) were infected. The difference between sexes was not significant (Chi-square test). Infected females were significantly larger than non-infected ones, but there was no difference in lengths between infected and non-infected males (Wilcoxon rank sum test). No significant differences were found in the length of infected fish when compared between sexes.

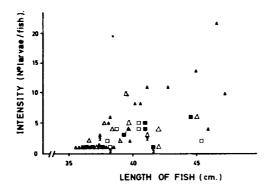


Fig. 1: Relationship between the length of Trachurus murphyi and the intensity of infections with Anisakis sp. larvae, according to sex and sexual maturity stage of the host. ( $\blacktriangle$ ) Mature females, ( $\bigtriangleup$ ) Immature females, ( $\blacksquare$ ) Mature males and ( $\Box$ ) Immature males.

Relación entre la longitud de *Trachurus murphyi* y la intensidad de las infecciones por larvas Anisakis sp., según el sexo y estado de madurez sexual del huésped. ( $\blacktriangle$ ) Hembras maduras, ( $\bigtriangleup$ ) hembras inmaduras, ( $\blacksquare$ ) machos maduros y ( $\square$ ) machos inmaduros.

Mean intensity of infections was not significantly different between sexes (Wilcoxon rank sum test; WRST), but variance was significatively greater among females (Miller Jackniffe test (MJT), k = 1; Q = 3.42 P < 0.001; Table 2). Intensities of infection were significantly correlated with length of infected fish in both sexes ( $r_s \ 9 = 0.62$ ; P < 0.01;  $r_s \ 0.49$ ; P < 0.05).

Sexual maturity stage seems to be an important factor influencing the infection levels in fish examined, because those hosts which were mature had a significatively higher prevalence of infection than had immature fish (Chi-square test (CST); P < 0.05). Although infected mature and immature fishes were not different in length, nor in mean intensity of infections (WRST), variance of parasite intensities was significatively higher among mature fishes (MJT, k = 1; Q = 1.67; P < 0.05). Thus, due to the skewed frequency distribution of intensities, highest parasite intensities occurred among mature fishes. When sex was considered separately, the same results in prevalence, (CST; P < 0.001) mean intensity of infections (WRST; P > 0.05), and variance of intensities (MJT, k = 1; Q = 1.65; P < 0.05) were obtained only for females, but not for males. Furthermore, mature females had a significantly higher prevalence of infection (CST; P < 0.01) and variance of parasite intensities than had mature males, (MJT; k = 1; Q = 3.05;

#### TABLE 2

Number of *T. murphyi* infected and uninfected with *Anisakis* sp. larvae, mean parasite intensity  $(\bar{x})$  and variance to mean ratio  $(S^2/\bar{x})$ , by sex and sexual maturity stage of the host.

Número de *T. murphyi* infectados y no infectados con larvas *Anisakis* sp., intensidad parasitaria promedio  $(\bar{x})$  y razón entre varianza y promedio  $(S^2/\bar{x})$ , según sexo y estado de madurez sexual del huésped.

	BOTH SEXES		FEMALES		MALES	
	Infected	Uninfected	Infected	Uninfected	Infected	Uninfected
Immature fish						
Total	23	39	15	29	8	10
x	3.19(21)*		3.62(13)*		2.50	
$S^2/x$	1.59		1.77		1.03	
Mature fish						
Total	31	27	21	8	10	19
x	4.70(30)*		5.62		2.56	
$\overline{x}$ S <sup>2</sup> /x	5.14		5.42		1.57	
All fish						
Total	54	66	36	37	18	29
x	4.08(51)*		4.85 (34)*		2.53(17)*	
$S^2/x$	4.07		4.49		1.24	

\* Number of fish considered for calculation of mean intensity.

\* Número de peces considerados para el cálculo de la intensidad media.

P < 0.01) whereas no differences were found among immature fishes when prevalence of infections, mean intensity of infections and their variances were compared between sexes (Table 2 and Figure 1).

Anisakis sp. larvae were distributed in the 54 infected fish as follows: 38 fish (70%) had parasites localized in only one site (27 fish were infected only in the gonadic mesenteries and 11 fish infected only in the visceral mesenteries) whereas 16 fish (30%) were infected simultaneously in several body locations. The most frequent locations of larvae are shown in Table 3.

# TABLE 3

Number of *T. murphyi* infected and total number of Anisakis sp. larvae collected in each body location.

Número de T. murphyi infectados y número total de larvas Anisakis sp. colectadas en cada localización corporal.

	Number of fish infected	Total number of parasites collected		
Visceral mesenteries	21	89		
Gonadic mesenteries	38	100		
Liver	3	3		
Muscles	3	5		
Other viscera	5	11		

Rank correlation between parasite intensities and Fulton's condition factor of infected fish (see Margolis 1970) showed a low but significant value between these variables ( $r_s = 0.18$ ; P < 0.05).

# DISCUSSION

Trachurus murphyi is the second most important marketable fish in Chile in relation to the volume of landings (Serra et al. 1979). This epipelagic fish feeds mainly on copepods, pteropods, euphausiids and clupeids (Videla, 1976), but the relative importance of these food items in the diet probably changes with geographic locality (see Rosario 1970). Probably decapods, euphausiids and clupeids transmit the parasites to T. murphyi, given that crustaceans have been reported as first intermediate hosts for Anisakis sp. larvae from North Atlantic. North Pacific and Japanese waters (Smith & Wootten 1978). Clupeids are claimed to be responsible for transmitting these larvae to Baltic cod (Gradba 1976). However, in T. murphyi no direct evidence exists to support these possible pathways of infection. Initial appearance of infections with Anisakis sp. larvae is probably related to a qualitative change in food habits of T. murphyi, but there is no information about its food habits at different ages.

Anisakis sp. larvae in jack mackerels of the genus Trachurus have been previously reported for T. symmetricus by Dailey (1969) in the North-eastern Pacific ocean (52% of fish infected). Japanese investigators have found a high rate of infection in T. japonicus (Smith & Wootten 1978). Tantaleán (1972) reported that 48% of T. murphyi caught along the Callao coast, Perú, were infected. Cattan & Videla (1976) found 28% of T. murphyi infected in a sample collected off the coast of Arica, Chile.

There is no previous information about the intensity of infections with Anisakis sp. larvae in T. murphyi. Our results show that intensity of infections are much lower than those reported for clupeids, gadoids and salmonids, probably due to a different predation pressure upon intermediate hosts. However, increasing intensity and prevalence of infections with length class as seen in our data, seems to be a worldwide pattern among species of fish which are host of Anisakis.

Differences between our results and those previously reported for jack mackerels could be due to the different abundances of intermediate and/or definitive hosts in the localities sampled, to differences in the age and/or length composition of fish in the sample, as well as to the year of sampling (Bishop & Margolis 1955, Khalil 1969, Davey 1972, Young 1972, Van Banning & Becker 1978).

We strongly believe that the almost absolute absence of reports on sex related differences in prevalence and/or intensity of infection with *Anisakis* sp. larvae in marine fish (exception made of Collard 1970 and Cattan & Videla 1976) is mainly due to the statistical procedures employed in the analysis of data. Generally data are log-(x + 1) transformed before comparisons between sexes are made. Although we recognize the good stabilizing properties of the log-(x + 1) transformation on the variance of intensities (made in order to

apply statistical methods based on the normal distribution), we think that the concomitant shortening of the tails of the frequency distribution of intensities causes serious problems. It not only underestimates the importance that the few heavily parasitized hosts have for the parasite population (Kennedy 1975), but also distorts the variance to mean ratio, which is a good indicator of the parasite clumping in the host population only when it is calculated with untransformed data. For example, when our data were transformed as described above, F-tests were unable to detect the differences in variances of intensities that untransformed data revealed.

Furthermore, the addition of one to the intensities, leads to the inclusion of the prevalence in the comparison, which results in a comparison of abundances (sensu Margolis et al. 1982) rather than in a comparison of mean intensities. If a significant difference were found, no one could know if it was due to differences in the prevalences or in the intensities.

We believe that there are good reasons to expect diferences in intensity and/or prevalence of infections between sexes of fish hosts of Anisakis sp. larvae. Differential growth rate between sexes or different energy requirements during gonad maturation could possibly help to explain these differences. The first reason (different growth rate) is not applicable to T. murphyi because sexes grow at similar rates (Serra et al. 1979), but the second is plausible, because females yearly build up gonads that reach a significantly lower gonosomatic index than do males (Mainguyague 1980). This suggests than females should have higher energy requirements than males, thus rendering the females more prone to infection because they need to ingest more food.

Location of parasites within the host body, mainly coelozoic, coincides with the generally reported distribution pattern for these larvae in other fish species host (Cheng 1976). The almost absolute absence of larvae in the muscles of *T. murphyi* not only indicates that there is little health hazard for human consumption, but also suggest differences from its distribution in clupeids, gadoids and salmonids where comparatively more larvae are found in the muscles (Novotny & Uzmann 1960, Wootten & Waddell 1977, Beverley-Burton & Pippy 1978, Van Banning & Becker 1978, Carvajal et al. 1979).

In T. murphyi there is a noticeable difference in the prevalence and intensity of liver infections in comparison to that found in the fish families referred to above. It is possible that chemical composition of the liver and flesh tissue of different fish host species, as well as the closeness of these organs to the stomach influences the distribution of parasites within the host body. The location pattern of Anisakis sp. larvae within T. murphyi probably accounts for the low correlation found between the condition factor of fish and parasite intensities, given that the major pathological effects occur in its liver and muscles (Margolis 1970, Cheng 1976).

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