

***Camallanus maculatus* n. sp. (Nematoda: Camallanidae) in an ornamental fish
Xiphophorus maculatus (Osteichthyes: Poeciliidae)
cultivated in São Paulo State, Brazil**

[*Camallanus maculatus* n. sp. (Nematoda: Camallanidae) em peixe ornamental *Xiphophorus maculatus* (Osteichthyes: Poeciliidae) cultivado no estado de São Paulo, Brasil]

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ABSTRACT

The present study describes *Camallanus maculatus* n. sp. (Nematoda: Camallanidae) from *Xiphophorus maculatus* (Osteichthyes: Poeciliidae) collected in an ornamental fish farm in São Paulo State, Brazil. Thirty-three out of 40 examined fish were parasitized (82% prevalence) with mean intensity of 2.8 (1-20). Male specimens were 3,500µm total length and females 7,100µm total length, provided with orange-brown buccal capsule and two developed tridents. Males showed two unequal spicules, gubernaculum absent and females with vulva situated in the middle of the body. The main characteristics were the sleekly longitudinal ridges in the buccal capsule in both sexes, enlargement at the anterior end of the smaller spicule, the number of genital papillae, longer muscular and glandular portion of esophagus and round tail in female.

Keywords: fish, *Xiphophorus maculatus*, nematode, prevalence, *Camallanus maculatus* n. sp.

RESUMO

Descreve-se *Camallanus maculatus* n. sp. (Nematoda: Camallanidae) em *Xiphophorus maculatus* (Osteichthyes: Poeciliidae) capturado em uma piscicultura ornamental no estado de São Paulo. De 40 peixes examinados, 33 estavam parasitados (prevalência de 82%) cuja intensidade média foi de 2,8 (1-20). Os machos apresentaram 3.500µm e as fêmeas com 7,100µm de comprimento total, providos de uma cápsula bucal laranja-castanho e dois tridentes. Observaram-se machos com espículos desiguais, gubernáculo ausente e fêmeas com vulva situada na metade do corpo. As principais características foram as estrias longitudinais lisas em cada valva da cápsula bucal em ambos os sexos, o espículo menor com alargamento na extremidade anterior, o número de papilas genitais, as porções muscular e glandular do esôfago longas e a cauda longa e arredondada na fêmea.

Palavras-chave: peixe, *Xiphophorus maculatus*, nematóide, prevalência, *Camallanus maculatus* n. sp.

INTRODUCTION

There is much profitable information concerning ornamental fish industry, one of them come from Evans and Lester (2001), who pointed out that it is estimated at 900 million dollars. National and international transport of fish contributes on

parasites dissemination not only in ornamental, but also in native fish.

The presence of parasites in several ornamental fish species is mentioned by (Moravec et al., 1999) in Germany, Australia (Evans and Lester, 2001), Korea (Kim et al., 2002) and Brazil (Martins et al., 2007). Nematodes of the genus *Camallanus* Railliet and Henry, 1915, are widely distributed over the

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world and, in Brazil, *Camallanus acaudatus* and *Camallanus tridentatus* were found in *Osteoglossum bicirrhosum* and *Arapaima gigas* by Ferraz and Thatcher (1990). They could be seen into reared and native fish intestines (Amin, 1978; Baker, 1979; Ferraz and Thatcher, 1990; Lakshimi et al., 1990; Moravec 1994; Torres et al., 1991; Moravec, 1994; Martins et al., 2007). Their prevalence may reach 83.3% for both salmonids (Torres et al., 1990) and *Poecilia reticulata* (Kim et al., 2002) and 2.6% in *Xiphophorus maculatus* from ornamental fish in Florianópolis-SC (Piazza et al., 2006). This ornamental fish has a great economical importance to Brazil, since it can be found anywhere and it is very well traded.

This study describes a new species of *Camallanus* found during a parasitological fauna survey of *Xiphophorus maculatus* Günther, 1866.

MATERIAL AND METHODS

Forty specimens of *X. maculatus* (Osteichthyes: Poeciliidae) weighting 0.50 ± 0.1 g were collected from an ornamental fish farm in Araraquara-SP, Brazil, and also examined to find out if there were any nematodes. As the answer was affirmative, the researchers removed the nematodes carefully from fish intestines to fix them in AFA, at 65°C for posterior preservation in 70% alcohol with 5% glycerine. Eighteen males and 23 females were studied after dehydration and cleared in Amann lactofenol, acetic acid or glycerine afterwards the drawings were prepared in camera lucida. Twenty larvae were measured from the studied females as well as identified according to Chabaud (1975) and Moravec (1973, 1994). Specimens deposited at the Helminthological Collection of Oswaldo Cruz Institute¹. Holotype male, allotype female and paratypes (CHIOC n. 35283).

RESULTS AND DISCUSSION

Figure 1 (1-10) shows details of a male of *Camallanus maculatus* n.sp. from *X. maculatus*.

In order to observe the morphology of medium sized nematodes, smooth cuticle and red colour in life were observed, but not cervical papillae. It

was also registered that their mouth opening was surrounded by two pairs of papillae and four sclerotic plates. The anterior end was provided with a large orange-brown buccal capsule with two sclerotic lateral valves, a short basal ring and two developed tridents, which slightly exceeded the basal ring. Internal surface of each valve, supported by sleekly longitudinal ridges, varied on numbers for both sexes. Each valve had a pair of brown thickenings on its anterior surface.

This trial also aimed at observing the oesophagus that is divided in two parts, the anterior and posterior muscular portions. From the analysis, the first one is slightly smaller than the second portion. There is another important issue, the opening of glandular oesophagus into intestine, provided by valves. Nevertheless, it was difficult to visualize the excretory pore, situated just after nerve ring that encircles the anterior portion of muscular oesophagus.

Males presented two different spicules, but their gubernaculum was absent. On the other hand, females had their vulvas in the middle of their bodies, indicating that they are viviparous (Fig. 1-10).

The mouth of the male had two posterior directed brown chitinous tridents, with three prongs each, described as a 90 ± 7 (80-100) μ m long middle prong of tridents and two lateral prongs that were 81 ± 3 (80-90) μ m long. Internal surface of valves was provided by 26 to 28 longitudinal ridges while spicules were different, simple and pointed to the posterior end. The small spicule was thin and provided by an enlargement. The caudal alae was narrowed as well as provided by pedunculated papillae with seven pairs of preanal and six pairs of postanal. The first three pairs were closed to one another as they were all together. The fifth pair of postanal papilla was larger than the others (see Fig. 1.8). According to the lateral view, when the cloacae region was surrounded, it could be seen two transversal structures, two pairs of sessile papillae (adanal) and a ventral flexed tail (Table 1).

Females also presented two posterior directed brown chitinous tridents, with three prongs each, described as a 135 ± 7 (130-140) μ m long middle prong of tridents and two lateral prongs that were 117 ± 10 (110-124) μ m long. The internal surface of valves was provided by 30 to 32 longitudinal ridges.

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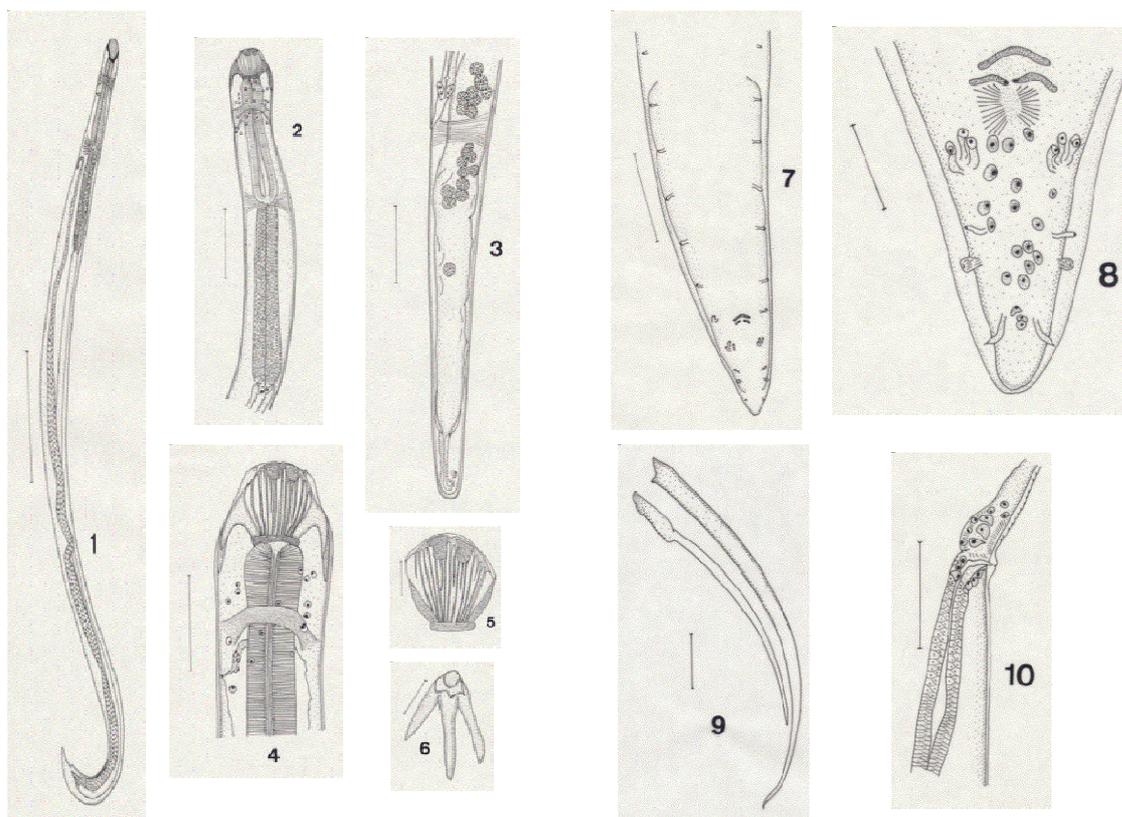


Figure 1. Male of *Camallanus maculatus* n. sp. from *Xiphophorus maculatus* (Bar=500µm). Figure 2. Anterior extremity of male showing oesophagus, lateral view (Bar=200µm). Figure 3. Lateral extremity of male, lateral view (Bar=200µm). Figure 4. Anterior extremity of male with detail of excretory pore, lateral view (Bar=100µm). Figure 5. Buccal capsule of female, lateral view (Bar=50µm). Figure 6. Trident of female, dorsal view (Bar=50µm).

Figure 7. Posterior end of male showing papillae and caudal alae, ventral view (Bar=100µm). Figure 8. Posterior end of male with details of the postanal papillae (note the shape of the fifth pair), ventral view (Bar=30µm). Figure 9. Spicules (Bar=20µm). Figure 10. Lateral view of vulva (Bar=100µm).

During the study of their reproductive organs, it was observed that muscular vagina was posterior directed from vulva. Vagina vera, measured in eight specimens, was 57 ± 6 (50-72)µm long and vagina uterine, from eight specimens, was 212 ± 23 (180-264)µm long. Uterus was posterior directed to the end of the tail. In order to measure their eggs, thirty of them presented 34 ± 2 (30-38)µm of diameter. From dissected females, the larvae into the uterus measured 390 ± 7 (380-400)µm long by 20 ± 2 (18-24)µm width as well as a rounded tail (Table 1).

The high prevalence rate from this study was similar to *Camallanus oxycephalus* Ward and Magath, 1916 (Stromberg and Crites, 1975), *Camallanus* sp. (Lyan and Seng, 1979), *Camallanus corderoi* Torres, Teuber and Miranda, 1990 (Torres et al., 1990) and *Camallanus cotti* Fujita, 1927 (Font and Tate, 1994; Rigby et al.,

1997; Kim et al., 2002). On the other hand, the mean intensity was higher than the one registered by Torres et al. (1991), in salmonids.

Aiming at comparing the described species, the nematodes were arranged according to the presence of tridents, two spicules and longitudinal ridges shape, as these main characteristics were suggested by Moravec (1973) and Gupta and Verma (1978). Measurements of both sexes were different in total length, buccal capsule length and width, number of longitudinal ridges, muscular and glandular oesophagus length, number of genital papillae and spicules length from *Camallanus longicaudatus* Moravec, 1973; *Camallanus kirandensis* Amin, 1978; *Camallanus ancyloides* Baker, 1979; *Camallanus tholukodensis* Lakshmi, Rao and Shyamasundari, 1990.

Camallanus maculatus n. sp...

The present specimens differ from *Camallanus lacustris* Zoega, 1776 (Moravec, 1994) due to the greater number of longitudinal ridges in buccal capsule, the highest measures of tridents in both sexes, the absence of mucrons on tails of females, shorter glandular oesophagus, nerve ring position

and longer tail in females. However, similar features were observed by analysing *Camallanus testudineusi* Gupta and Verma, 1978, *C. cotti* (Moravec and Sey, 1988; Rigby et al., 1997); *C. corderoi* (Torres et al., 1990) and *Camallanus truncatus* Rudolphi, 1814 (Moravec, 1971, 1994).

Table 1. Measurements of *Camallanus maculatus* from *Xiphophorus maculatus* in São Paulo State, Brazil

Characteristic or features	Male	Female
Total length	3,500±363 (2,700-4,100)	7,100±1461 (4,400-9,020)
Width	168±31 (80-204)	298±73 (163-450)
Buccal capsule L	79±4 (70-84)	131±43 (110-320)
Buccal capsule W	80±9 (70-96)	137±43 (116-324)
Basal ring L	11±5 (4-20)	16±7 (4-40)
Basal ring W	52±5 (40-60)	75±16 (20-100)
Muscular Oesophagus L	342±49 (256-400)	521±60 (440-656)
Muscular Oesophagus W	82±16 (40-120)	91±11 (72-120)
Glandular Oesophagus L	452±54 (392-560)	610±54 (496-680)
Glandular Oesophagus W	78±15 (32-96)	95±15 (64-120)
Nerve ring ¹	141±41 (80-200)	236±44 (120-280)
Excretory pore ¹	250±42 (240-270)	380±72 (360-400)
Large spicule L	148±23 (107-204)	-
Large spicule W	11±2 (8-16)	-
Small spicule L	99±11 (80-124)	-
Small spicule W	6±2 (3-10)	-
Caudal alae L	323±57 (230-390)	-
Caudal alae W	12±4 (4-18)	-
Testis L	3,000±390 (2,400-3,900)	-
Testis W	71±17 (40-96)	-
Testis reflexion ²	665±203 (448-1,040)	-
Vulva ¹	-	3,900±856 (2,500-5,100)
Tail	115±8 (92-120)	900±180 (700-1,300)

L: length; W: width; ¹Distance of the structure from anterior extremity; ²Distance of the testis reflexion to the anterior extremity. Averages are in micrometers followed by standard deviation and range in parentheses

The present nematodes may be distinguished from *C. testudineusi* by a greater number of longitudinal ridges and their sleekly appearances, smaller buccal capsule and basal ring, shorter spicules, number of genital papillae and also by an enlargement at the anterior end of the smaller spicule. Also, they differ from *C. cotti* (Moravec and Sey, 1988; Rigby et al., 1997) by nerve ring position, testis flexure more distant from anterior end, shorter caudal alae, the highest measures of spicules in males and once more by an enlargement at the anterior extremity of the smaller spicule. Moreover, the present specimens can also be distinguished by their greater number of longitudinal ridges, excretory pore position and trident prongs, which are longer than those ones observed in *C. cotti* in both sexes.

Although *C. truncatus*, studied by Moravec (1971, 1994), showed similar measurements of body length, buccal capsule, nerve ring position, spicules length and number of genital papillae in males, some strong differences must be inferred. Male specimens here described presented significantly increased number of longitudinal ridges, shorter muscular and glandular oesophagus, and the shape of the smaller spicule that has an enlargement at the anterior end. Otherwise, measurements of females showed larger buccal capsule and tridents, shorter muscular and glandular oesophagus, the shortest distance of vulva to anterior end and longer tail. While in *C. truncatus*, trident prongs exceeded greatly on the posterior edge of buccal capsule, in this study, the prongs surpassed the posterior edge of buccal capsule moderately.

Despite the similarity of the present material with *C. anabantis* measurements from De and Ghosh, 1989, important characteristics as smaller spicules length and shape, absence of mucrons in male, female tail and number of genital papillae were the main differences. By analyses, *C. kilasirii* De and Ghosh, 1989 and *C. carangis* Rigby, Adamson and Deardorff, 1998, found out that these nematodes differed not only due to mucrons absence, but also because of genital papillae number, the position of nerve ring and excretory pore, the shape and length of spicules and longitudinal ridges in buccal capsule.

The present description resembles *C. corderoi* (Torres et al., 1990) to body length, buccal capsule, basal ring, smaller spicule in males, number and shape of longitudinal ridges. By the fact that *C. corderoi* was the most similar species, it could be observed the original material from *Percichthys trucha* from Villarica Lake, in Chile, kindly donated by Torres et al. (1990). The strongest differences were the shape of longitudinal ridges, smaller spicule and tail shapes of females (Fig. 11, B-D). Males and females of *C. corderoi* had thick rings along the longitudinal ridges, nevertheless, the studied specimens here presented a sleek structure (Fig. 11, A-C).

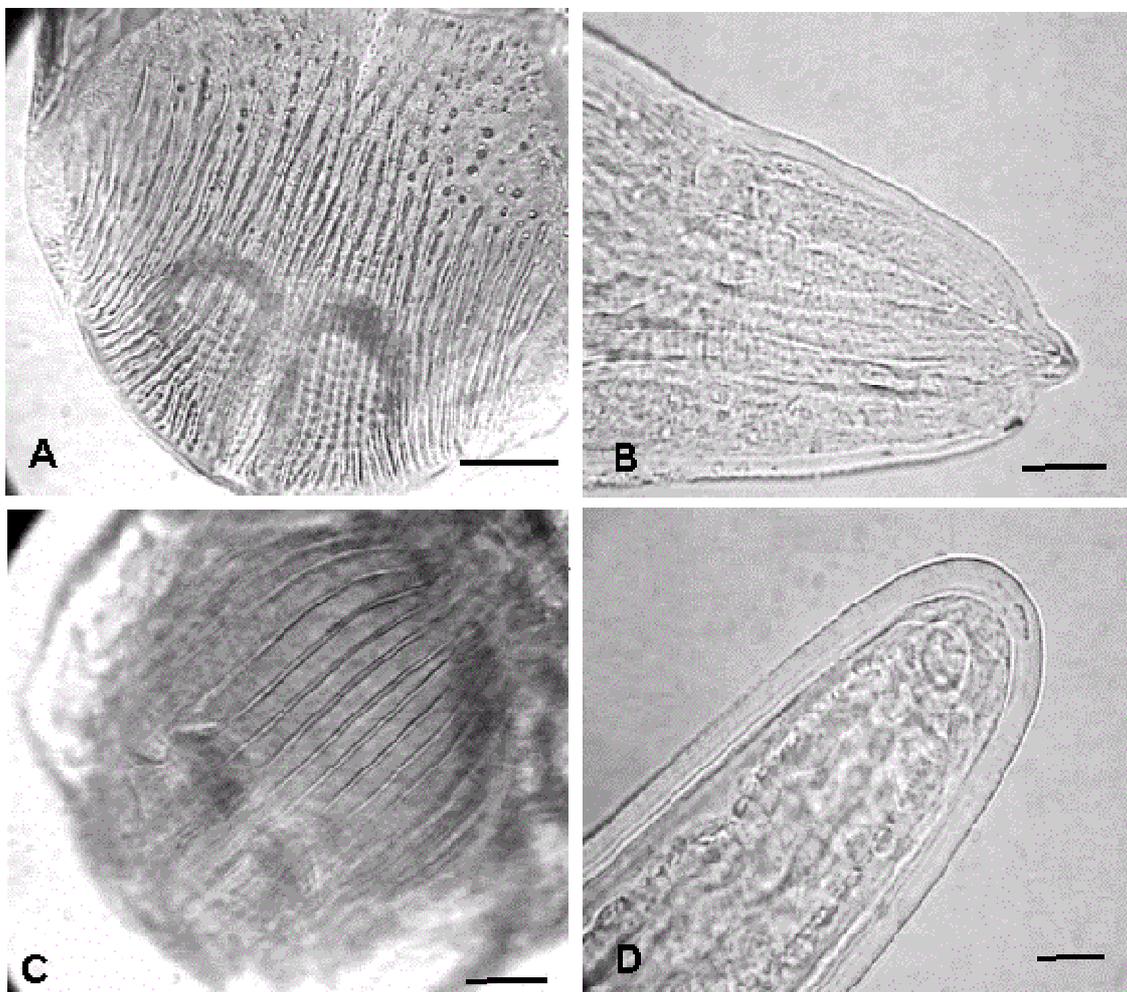


Figure 11. Shape of the longitudinal ridges in the buccal capsule and female tail of *Camallanus corderoi* Torres, Teuber and Miranda, 1990 from *Percichthys trucha* (A-B) and *Camallanus maculatus* n. sp. from *Xiphophorus maculatus* (C-D). Bar=20µm.

The smaller spicule had an enlargement in its anterior end and the tail end is longer and in the females rounded when compared to shorter and conical tail of *C. corderoi*. Other results regarding spicule length showed significantly shorter dimensions when compared to those already shown in other researches. Once more, it differed from *C. corderoi* by a shorter muscular and glandular portion of oesophagus that was easily confirmed by analysing those specimens. Finally, the present description differed from *Camallanus acaudatus* Ferraz and Thatcher, 1990, described in Brazil, by the presence of two spicules, the highest measurements of body length, buccal capsule, glandular oesophagus, excretory pore position, significantly shorter caudal alae, as well as number of genital papillae and tail in males. When compared to *Camallanus tridentatus* Drasche, 1884 (Ferraz and Thatcher, 1990), these specimens had shown larger buccal capsule, greater number of longitudinal ridges, number of post-anal papillae and two spicules in males. Almost all measurements differed, but basal ring and excretory pore in *C. acaudatus* and *C. tridentatus* females. Important differences, that must be commented, are the sub-terminal anus in *C. acaudatus* and bifurcated tail in *C. tridentatus* females, which were very different from this description.

At last, by the differences observed in buccal capsule concerning longitudinal ridges and smaller spicule shapes, excretory pore, nerve ring position, muscular, glandular oesophagus, spicules length, number of genital papillae, caudal alae length in males and several characteristics of females such as their tails end, the authors suggest that there is a new species and a new host for camallanid nematode in Brazil.

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