

# ORIGINAL ARTICLE /ARTÍCULO ORIGINAL

LERNAEOSIS IN CNESTERODON DECEMMACULATUS (CYPRINODONTIFORMES, POECILIDAE) AND OBSERVATIONS ON THE LETHAL EFFECT IN SMALL SIZED SPECIES OF HOSTS

LERNEOSIS EN CNESTERODON DECEMMACULATUS (CYPRINODONTIFORMES, POECILIDAE) Y OBSERVACIONES SOBRE EL EFECTO LETAL SOBRE ESPECIES HOSPEDADORAS DE PEQUEÑA TALLA

Silvia Guagliardo & Daniel Tanzola

Laboratorio de Patología de Organismos Acuáticos de Interés Comercial. Universidad Nacional del Sur, Bahía Blanca, Argentina. E-mail: sguaglia@criba.edu.ar

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### **ABSTRACT**

Lernaea cyprinacea is one of the best known species of the genus, with a wide global distribution, that causes lesions on the skin of fresh water fish fostering secondary infections. The aim of the present study was to record the presence of *L. cyprinacea* on *Cnesterodon decemmaculatus* and to study the lethal effect of this copepod on a host species of small size. The specimens were parasitized by post-metamorphic stages of females. The ulcerous lesions rapidly became contaminated with dense *Saprolegnia* sp. mycelia and they were associated with *Epistylis* sp., a ciliated peritrichid. Histological examination showed invasion of the visceral mass which exerted mechanical compression on the muscle, kidney, spleen and gonad. Fibrous encapsulation and inflammatory infiltrate were observed as a response. The intense visceral invasion provoked in *C. decemmaculatus* in the reproductive stages makes the fish potentially susceptible to lethal consequences, with consequent damage at the population level.

Keywords: Lernaea cyprinacea – Cnesterodon decemmaculatus – Fish pathology

#### RESUMEN

Lernaea cyprinacea es una de las especies mejor conocidas del género, con una distribución mundial, que causa lesiones en la piel de los peces de agua dulce promoviendo infecciones secundarias. El objetivo del presente estudio fue registrar a L. cyrpinacea en Cnesterodon decemmaculatus y estudiar el efecto letal de éste copépodo en una especie de hospedador de pequeña talla. Los especímenes estuvieron parasitados por hembras post-metamórficas. Las lesiones ulcerativas rápudamente se contaminaron con densos micelios de Saprolegnia sp. asociados con el ciliado peritrichido Epistylis sp. El examen histológico mostró invasión de la masa visceral el cual ejerció compression mecánica sobre el músculo, el riñón, el bazo y las gónadas. Se observó una respuesta de encapsulación fibrosa e infiltrado inflamatorio. La intensa invasión visceral de C. decemmaculatus en estados reproductivos torna al pez potencialmente susceptible a consecuencias letales con el perjuicio consecutivo a nivel poblacional.

Palabras clave: Lernaea cyprinacea - Cnesterodon decemmaculatus - Patología de peces

## **INTRODUCTION**

#### MATERIALS AND METHOD

Cnesterodon decemmaculatus, Jenyns, 1842, is one of the small species of the Argentine ichthyofauna. They live in shallow pools and streams from the south of Brazil to Río Negro (Lucinda, 2005). Until now it was not known as a host to Lernaea cyprinacea Linnaeus, 1758 (Copepoda, Cyclopoida), a species introduced accidentally in South America via the Cyprinids (Piasecki et al., 2004). Records of anchor worms in Argentina are occasional (Paggi, 1972, 1976; Ortubay et al., 1994; Roux et al., 2000; Vanotti and Tanzola, 2005, Mancini et al., 2006, 2008; Plaul et al., 2010). Lernaea cyprinacea is one of the best known species in the genus, with the widest global distribution, that causes skin lesions on fresh water fish. Cases have been reported of secondary infections in unsuitable water conditions (Carnevia & Speranza, 2003; Mancini et al., 2008, Bednarska et al., 2009). Among the lesions caused by different species of Lernaea the following are mentioned: disruption of the dermis and epidermis, hyperplasia, acute and chronic inflammation, granuloma formation, fibrotic encapsulation, ulceration and necrosis, and the following cell types of the immune response are observed: neutrophils, lymphocytes and eosinophilic granule cells (ECGs). Fish mortality appears to be related to the size of the fish (Sharif & Roberts, 1989; Paperna, 1996; Mancini et al., 2008).

The objective of the present study is to record the presence of *L. cyprinacea* in *C. decemmaculatus* and to study the lethal effect of this copepod on a small sized host species. They have not experimented with live animals, not affected population parameters of the host or provoked impacts on the environment and human health or animal.

One hundred and eighteen adults of Cnesterodon decemmaculatus (62 females and 56 males) from an urban lake in Bahía Blanca, Argentina (38°41'S; 62°15'W) were examined during March and April 2012. They were between 23-34 mm (Mean= 28; Standard deviation= 3.02) in size and had a total weight between 0.227-0.277 g (Mean= 0.250; Standard deviation= 0.018). A hand net was used to catch the fish which were then kept in an aerated aquarium at 18-20°C and fed with fish food. The external areas of the body examined were: skin, eyes and fins. The parasitized fish were fixed in 10% formalin, transverse sections in series (5 µm thick) were made of the whole body and stained with hematoxylin and eosin (HE) and Masson trichrome.

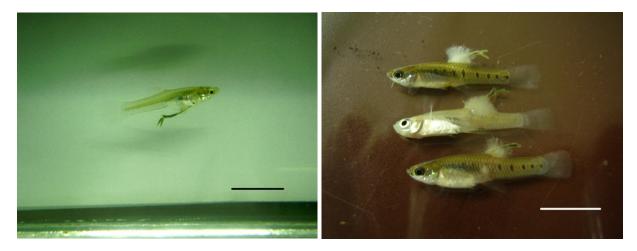
## **RESULTS**

The specimens were parasitized by postmetamorphic stages of females of L. cyprinacea. They were identified by morphological features such as the structure of antennae, maxillae, maxillipeds and thoracopods. The site of anchorage of the copepods varied between dorsal (8/19), anal (9/19) and left pectoral fins (2/19) (Figures 1a and 1b). The prevalence of infection was 19.1% and the mean intensity 1. Ulcerative lesions were rapidly contaminated with dense mycelia of Saprolegnia sp. that invaded the external integument and the scale sack. Dense colonies of *Epistylis* sp (ciliated peritrichids) and Gomphonema spp (diatoms) were fixed to the external segment of the neck of L. cyprinacea.

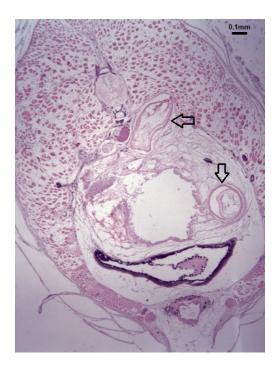
The histological examination showed that the whole visceral mass of the fish was invaded. Figures 2 illustrate the anchor apparatus

(arrows) of the parasite invading internal organs and exerting mechanical compression on the muscle and kidney. The fish developed a strong fibrotic response around the horns of the anchor. Figure 3 shows infiltrated inflammatory tissue around the parasite, severe compressive atrophy in the spleen. No

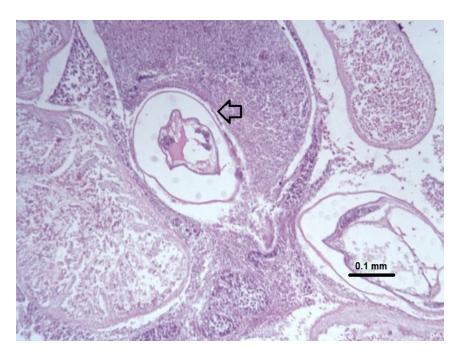
congestion or hemorrhagic areas in the visceral mass were observed. All the parasitized fish became emaciated and died at the end of three weeks. In contrast, there was 100% survival in another fish group without parasites maintained in the same conditions.



**Figure 1.** Lernaea cyprinacea (in situ) parasitising Cnesterodon decemmaculatus. a) Ventral location of anchorage; b) Dorsal fin affected, observed the heavy colonization by micelia of Saprolegnia sp., Epystilis sp. and Gomphonema sp. (Scale-bar=10mm)



**Figure 2.** Transverse section of *Cnesterodon decemmaculatus* showing two sections (arrows) of the anchorage aparatus (horns) exerting mechanical compression on the muscle and kidney and a strong fibrosis arround the visceral mass (Scale-bar=0.1mm)



**Figure 3.** Transverse section of *Cnesterodon decemmaculatus* showing severe compressive atrophy in the spleen (arrow) (Scalebar=0.1mm)

## **DISCUSSION**

Lernaea cyprinacea is a cosmopolitan species and one of the freshwater fish ectoparasites with lower host specificity. To the present, it has been recorded in 16 fish species in Argentina (Mancini et al., 2008; Plaul et al., 2010). This is the first record of *L. cyprinacea* in C. decemmaculatus. All parasitic females were ripped, coincident with the season of previous records in the literature (end of summer). In respect to the site of location on the host, the following points of insertion were reported in the literature: the base of the dorsal and pectoral fins and the middle part of the body (in Odontesthes bonariensis (Valenciennes, 1835)), the caudal peduncle (in Rhamdia quelen (Quoy & Gaimard, 1824)) or without any preferred site in Oligosarcus jenynsii (Günther, 1864) (Mancini et al., 2008). In the present study the invaded zones were the base of the dorsal, anal and left pectoral fins. Only one copepod was found per

fish, which appears to be the maximum permitted number considering the body mass ratio between parasite and host.

El-Mansy (2009) showed that specimens of Carassius auratus Linnaeus, 1758 infected by Lernaea were stressed, exhausted, lost scales and stopped feeding. They showed oedema in the anchorage point, hemorrhage, fibrotic reaction around the parasite, leukocyte infiltration, necrosis and ulcerative areas especially on the base of the fins. There are numerous reports of the presence of eosinophilic granular cells (EGCs), but it is still not clear what role they play in the inflammatory processes (Sharif & Roberts, 1989; Silva-Souza et al., 2000; Wojciech et al., 2004). In the present study, mononuclear infiltrates predominated around the parasite. Essa et al. (2003) demonstrated the deleterious effect of increased cortisol levels in the grass carp, Ctenopharyngodon idella Steindachner, 1866 parasitized by *L. cyprinacea*.

Sometimes infected fish were noted scratching on the walls of the tank (Sharif & Roberts, 1989). In the hosts studied in captivity a progressive loss of condition and positive response to feeding was observed. After three weeks 100% lethality was recorded compared to another group kept without parasites.

Considering the parasitic prevalence (P) and intensities (I) documented for Argentine fish, the estimates in this study can be classified as moderated (P=19%; I= 1) compared to 53.6% in O. bonariensis; 80.9% in O. jenynsii and 100% in Cyprinus carpio, among others. Paperna (1996) indicated that lernaeosis in fish smaller than 40 mm is lethal. The heavy invasion in the visceral mass of C. decemmaculatus makes the fish potentially susceptible of suffering lethal consequences in the reproductive stage, with consequent negative effect at population level, considering that the standard length recorded for this species varies between 38 mm (Ringuelet et al., 1967), and 35 mm (males) and 45 mm (females) (Lucinda, 2003).

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