FINDING OF THE NEMATODE LARVAE IN PREDATORY FISH SPECIES

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NALAZ LARVI NEMATODA KOD GRABLJIVIH VRSTA RIBA

Apstrakt

Neamatode iz roda *Eustrongylides* su paraziti ribojedih ptica. Njihov razvojni ciklus obuhvata dva prelazna domaćina, akvatične oligohete i planktofagne i bentofagne ribe npr. *Fundulus* ili *Gambosa, Neogobius,* ali i neke vrste iz porodice Cyprinidae koje kada pojedu inficiranu oligohetu postaju drugi prelazni domaćin. Grabljive vrste riba, kakav je som i smuđ, kada pojedu inficiranu ribu postaju paratenični domaćini dok njih ne pojedu ptice. Ovakvu ulogu mogu imati i vodozemci i gmizavci. Ustanovljeno je da ovi paraziti imaju uspešnu strategiju razvoja, s obzirom da jaje ostaje infektivno i do dve godine u spoljnoj sredini, a u prelaznom domaćinu može biti i preko godinu dana. Čovek nije tipičan domaćin ali se može zaraziti ukoliko jede sirovo ili nedovoljno termički obrađeno meso riba. U ovom radu prikazane su nematode koje se javljaju kod grabljivih slatkovodnih riba u Republici Srbiji. Istraživanja su sprovedena u periodu od 2011-2012 godine na kanalu Dunav-Tisza-Dunav, u gradskom području Novog Sada. Prikupljen je 21 uzorak smuđa (*Sander lucioperca*) težine od 250-500 g i 52 uzorka soma (*Siluris glanis*) težine 250-450 g. Urađen je postmortalni pregled abdominalne duplje, digestivnog trakta i drugih visceralnih organa. Utvrđeno je prisustvo nematoda kod 4 jedinke smuđa i 6 jedinki soma, što predstavlja prevalencu od 14.26%, odnosno 11.54%. Larve su bile prisutne u abdomenu, muskulaturi, lumenu želuca i želudačnom zidu gde su paraziti bili inkapsulirani. Broj parazita po ribi kretao se od nekoliko pa sve do 256. Sakupljene nematode fiksirane su u 70% etanol. Nakon fiksacije, svaka nematoda prosvetljivana je u mlečnoj kiselini radi morfološke observacije i identifikacije vrste. Relativni morfometrijski parametri i identifikacija parazita sprovedeni su prema ključevima Bauera (1987), Moraveca (1994) i Andersona (2000). Nematode su identifikovane kao *Eustrongylides* sp.- larveni oblici. Larve su bile crvenkaste boje. Dužina tela

larvi se kretala od 27 – 60.5 mm, a širina od 0.49 – 0.58 mm. Karakteriše ih prisustvo 12 papila na prednjem kraju, respoređenih u dva kruga po 6. Papile unutrašnjeg kruga su nešto izduženije Uzorci za patohistološki pregled uzeti su iz mišića i nodula koji su se nalazili u želudačnom zidu. Uzorci su bojeni standardnom metodom koristeći H&E. Paraziti konzumirani kod sveže i termički slabo obrađene ribe mogu predstavljati značajan rizik za ljude. Sveže meso ribe i tradicionalni riblji proizvodi pre nego što se nađu u prometu moraju biti pregledani na prisustvo nematoda. Adekvatna priprema ribljeg mesa jedna od najvažnijih mera opreza kao i podizanje javne svesti prilikom konzumacije ovih riba.

Ključne reči: Eustrongylides, nematode, predatorske ribe, larve
Keywords: Eustrongylides, nematodes, predatory fish, larvae

INTRODUCTION

Species from genus *Eustrongylides* have complex life cycles involving a definitive host and two intermediate hosts. Definitive hosts include aquatic birds mostly from order Ciconiiformes family Ardeidae, Anseriformes, Gaviiformes and Pelecaniformes (Spalding and Forrester 1993, Measures 1988). First intermediate hosts for *Eustrongylides sp*. are aquatic oligochaetes (Spalding et al. 1993). Second intermediate hosts are planktivorous and benthivorous fishes that could pass the infection on to fishes (paratenic hosts) and finally on to fish-eating birds (Moravec, 1994). Such exposure is usually common in larger fish species, like channel catfish - *Ictalurus punctatus* or pikeperch - *Sander lucioperca*, which, as predators, become infected with *Eustrongylides sp.* nematodes. In fish, these parasites are conspicuous as long, red, coiled individuals located in the body cavity or embedded in the muscle (Mitchum 1995, Overstreet 2003). They can produce grossly visible swelling or abdominal distention but mortalities from such infections are rarely reported and when they occur they usually involve secondary infections or suboptimal environmental conditions (Bursey 1982, Overstreet 2003).

In humans who have consumed raw or undercooked fish, *Eustrongylides sp.* have produced gastritis and intestinal perforation (Deardorff and Overstreet 1991; Cole 1999). Guerin et al. (1982) were the first to report a natural (accidental) human infection with *Eustrongylides sp.* The goal of this paper is to distinguish the presence of these types of nematodes in freshwater fish species in inland waters of Serbia and to indicate the need for adequate preparation of fish meat.

MATERIALS AND METHODS

Diagnostics and investigations were conducted in 2011-2012. Fifty two fish samples of European catfish *(Siluris glanis)* weighing 250-450 g and twenty one samples of zander (*Sander lucioperca*) were collected from eight locations on Danube-Tisza-Danube Canal. Fishes were parasitologically examined through cutting the body and the abdominal part, digestive tract and other ventral organs. Each fish was cut carefully from around the pectoral to the cloaca using scissors to observe the body cavity and to extract the viscera. All collected nematodes were were fixed in 70% ethanol for 24 h. After fixing, each nematode was cut at the anterior and posterior part of the body using a clean cover glass. Both ends of the body were cleared in lactic acid for morphological observation.

Relative parametars were measured and identification was performed using Bauer (1987), Moravec (1994) and Anderson (2000) keys. Samples for patohistological examination were taken from muscles and nodules in stomach walls. Samples were fixed in 10% water-buffered formalin and imbedded into paraffin blocks. 5μm thick slices were cut and stained with standard haematoxylin & eosin staining.

RESULTS AND DISCUSION

Presence of nematodes in the abdominal cavity (Figure 1), musculature (Figure 2), in the lumen of the stomach and encapsulated in stomach wall was revealed in 4 individuals of zander and 6 individuals of European catfish, what represented the prevalence of 14.26%, respectively 11.54%. The number of parasites per fish ranged from a few up to the 256. Parasites were determined as *Eustrongylides sp.* - larval form. This larva, was robust and pinkish red. Length of body was 27 – 60.5 mm, maximum width 0.49 – 0.58 mm. Buccal cavity 0.09 – 0.11 mm, oesophagus 0.907 – 1.35 mm. The larva retained a cuticle, and its twelve visible circle cephalic papillae were well defined. The number of papillae appeared to be six in each of two circles. The number of papillae was confirmed by apical observation with light microscopy. Inner circle papillae were spine-like apices and those of the outer circle were nipple-like apices. Patohistological observation of stomach and muscles are presented in Figure 3 and 4.

Figure 1. *Eusrongylides* sp. in the abdominal cavity of European catfish (*Siluris glanis*)

Figure 2. *Eusrongylides* sp. in the muscle of European catfish (*Siluris glanis*)

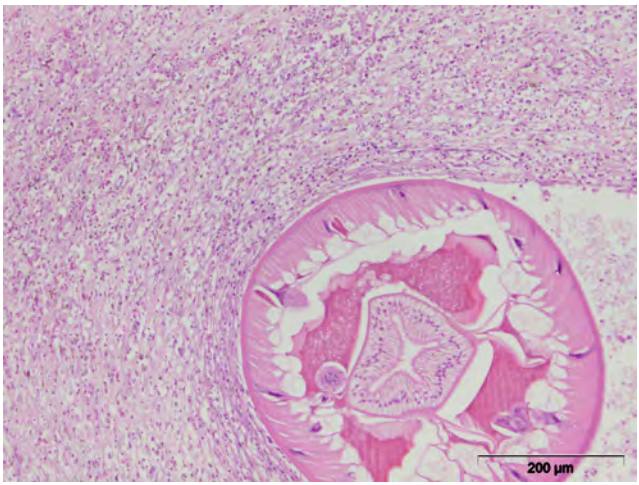

Figure 3. Cross-section of European catfish (*Siluris glanis*) stomach wall with *Eustrongylides* sp. larva (H&E).

Figure 4. Cross-section of European catfish (*Siluris glanis*) muscles with *Eustrongylides* sp. larva (H&E).

Fish parasites such as *Eustrongylides sp.* are highly important because they are capable to infect carnivorous organisms and humans who feed on them (Mohammad et al., 2011). Murrell (2002) suggested several control measures for preventing parasitic infections originating from freshwater, such as environmental control of surface water, hygienic aquaculture, and the control or elimination of the first intermediate hosts. FDA (2001) indicated that the effective methods for killing parasites are freezing, heating, adequate combination of salt content and storage time or hot smoking. On the other hand, brining and cold smoking may reduce the parasite hazard in fish, but they do not eliminate or minimize it to an acceptable level (Murrell, 2002). While health education is a key factor in combating zoonotic infections, experience in various countries has shown that for successful implementation of control measures, it is necessary, as Hughes (1992) points out, to have formal and informal cooperation between medical and veterinary interests at all levels of government, and with the community. According to Okumura et al. (1999) and Chieffi et al. (1992), the recommendation to avoid consumption of raw or poorly cooked fish is still the best preventive procedure.

CONCLUSIONS

Zoonotic nematodes can lead to human infection and special attention should be paid on them. Fresh fish meat and traditional fish product should be subjected to a visual examination for the purpose of detecting visible nematodes and other parasites before being placed in the trade. The consumption forms and the preparation of the fish food should be modified in a way that hazards to human health due to zoonotic parasites could be avoided. Health education is a key factor in combating zoonotic infections.

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