

## SHORT COMMUNICATION

### Third stage larvae of *Contracaecum* species in catfish (*Clarias gariepinus*) from Gaborone dam, Botswana

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#### ABSTRACT

A study was conducted with the aim of establishing the type of internal parasites in catfish (*Clarias gariepinus*) as well as the infection intensity. Ten catfish (*Clarias gariepinus*) specimens collected from the Gaborone Dam in Botswana were examined for internal parasites in August to November 2007. Eighty percent (80%) of these fish were found to be infected with third-stage larvae of *Contracaecum* spp in the abdominal cavity. The intensity of the infection was 14 to 250 worms per fish (mean intensity =  $60.3 \pm 9.70$ ). The mean length of third-stage larvae was  $14.25 \pm 3.28$  to  $35.88 \pm 4.96$  with a range of 10 to 42mm. No significant difference between host size and prevalence was established. *Contracaecum* species an anisakine nematode is potentially zoonotic. The public health implications of infected catfish are discussed.

**Keywords:** Catfish, larvae of *Contracaecum* spp, abdominal cavity, Botswana

#### INTRODUCTION

Gaborone dam, near Gaborone city, Botswana provides drinking water for the city's inhabitants and the environs as well as wild life and is also used for recreational fishing. There is prolific information on parasites of fresh water fish in Africa (Khalil, 1969; Aloo, 2001; Boomker, 1982; Chivasha, 1991; Douellou, 1992; Malvestuto and Ogambo-Ongoma, 1978; Barson, 2004; Barson and Avenant-Oldewage, 2006; Opara and Okon, 2002). There is a paucity of information on the parasites found in fish in Botswana.

*Contracaecum* species (Anisakidi) is a cosmopolitan parasite found as a parasite in piscivorous birds and mammals and can reach alarming intensities without killing the host (Hartwich, 1974; Lymbery *et al.*, 2002). In South Africa, *Contracaecum* species is one of the most prevalent parasites in piscoid migratory birds (Barson and Avenant - Oldewage, 2006). Fresh water fish such as the catfish (*Clarias gariepinus*) serve as definitive, intermediate or paratenic hosts in the life cycles of the widespread nematodes of the family Anisakidae parasites (Lymbery

*et al.*, 2002.). The life cycle of anisakine nematodes involving fish has been described previously (Whitfield and Hegg, 1977; Koic, M. and Fagerholm, 1995). Although less invasive than nematodes of the genera *Anisakis* and *Pseudoterranova* larvae of *Contracaecum* species have experimentally been shown to be infective to mammals (Vidal-Martinez *et al.*, 1994). Occasionally, third larval stage has been identified in people (Maroaka *et al.*, 1996; Lopez-Serrano, 2002; Noh *et al.*, 2003; Ito *et al.*, 2007).

In Botswana catfish, *C. gariepinus* (Burché) is a popular fish which is eaten by many people for it is relatively cheap, readily available from rivers and dams and regarded as a delicacy among fish eating communities in the city. The purpose of this investigation was to screen the catfish harvested from Gaborone dam for parasitic infections.

#### MATERIALS AND METHODS

All the catfish used in this study were purchased as fresh specimens from fish vendors who caught fish from Gaborone dam in the period between August to November, 2007. Usually two or three fish were collected

each month. The dam lies in a flat valley between Kgale and Oodi hills on the Notwane river situated on the South eastern corner of Botswana, 15 km from South African border post of Tlokweng. Whole fish were transported on ice to the laboratory for investigation.

### Collection of parasites

At the laboratory, the weight (g) of whole fish and standard length (cm) from head to the tail fin as well as sexes were determined. Using a pair of Scissors, a ventral incision was made and the mesenteric cavity was inspected for the presence of parasites. The visceral organs: namely, stomach, intestinal tract and liver as well as the body cavity and gills were opened and inspected for the presence of parasites separately. These were thoroughly washed with physiological saline. The washings were heated to 60°C in a water bath after which they were sieved (38 µm aperture). The residue was preserved in 4% (v/v) formal saline and checked for any parasites.

Larvae were counted, their anatomical sites of isolation documented. Thereafter, they were and then stored in 4% (v/v) formal saline for fixation. The specimens were cleared with glycerine before examination. The gastrointestinal tract from the oesophagus to the rectum was dissected and any worms encountered carefully detached from the stomach and intestinal mucosa by scraping with a knife or glass slide and examined microscopically for any parasites. Fillets were bluntly dissected and examined under white light for parasites. Internal organs such as the liver and kidneys were also checked for nematodes. All parasites isolated from the samples were recorded. Parasite taxonomic identification was based on gross morphometric and light microscopical observations. Results of the latter were compared with those of previous workers (Olson et al., 1983; Lymbery et al., 2002). The parasites were identified as shown by Yamaguti (1959 and 1961); Paperna (1996); Chabaud, (1974). Prevalence of larval nematodes (percentage of fish infected) and mean intensity (mean number of parasites per

infected fish) were subsequently calculated, as described by Olson et al., (1983)

Parasite body lengths were measured using a pair of Vernier calipers.

### Statistical analysis

Analysis of variance (ANOVA) statistics,  $X^2$  test and students t-test were performed (Gomez and Gomez, 1984).

## RESULTS AND DISCUSSION

In the present study, only one genus of anisakid larval nematode, *Contracaecum* spp was identified in catfish obtained from fresh water. Worms measuring 20–30.0 mm in body length and 0.3-0.4 mm diameter and positively identified as the third stage larvae of *Contracaecum* species were recovered from eight out of ten (80%) catfish (Table 1). This is the first report of an infection of the catfish with the third stage larval nematode, *Contracaecum* species in Botswana. Morphometric description of the isolated L3 larvae conformed to *Contracaecum* species as described by Cannon, 1977. The arithmetic average length and weight of male catfish were higher than that of the females. However, there was no sex predilection regarding infection intensity by the larval nematode *Contracaecum* spp. ( $X^2 = 2.2$   $P > 0.05$ ). It was surprising not to find other nematodes. Other workers have only been able to collect two nematodes from catfish. Barson and Avenant-Oldewage (2006) found a nematode, *Procamallanus larviconchus* in the stomach and larvae of *Contracaecum* spp in the abdominal cavity of catfish collected from Rietvlei Dam in South Africa. We are unable to account for the failure to find other nematodes and cestodes in the catfish from Gaborone dam.

The proportion of infected fish was 80% but 100% infection levels have been reported with high intensities of 700-2000 worms per fish (Mashego and Saayman, 1981; Boomker, 1982). This makes *Contracaecum* spp one of the most prevalent fish parasites in southern Africa. The high infection rate may be explained by the feeding habits of catfish. Major foods for catfish include aquatic

insects, crayfish, molluscs, crustaceans and other fishes. Crustaceans are intermediate or paratenic host for Anisakidae (Anderson, 1992). The presence of large numbers of 3<sup>rd</sup> stage larvae is attributed to the constant intake of small embryonated eggst. All the catfish examined were large.

Table 1. Parameters of catfish collected from Gaborone dam infected by *Contracaecum* species larva

Fish number	Sex	Length (cm)	Weight (Gram)	Number <i>Contracaecum</i> Species third larval stage (L3)
1	Male	59	1500	95
2	Female	60	2000	60
3	Male	87	2200	250
4	Male	58	2200	0
5	Female	54	1800	17
6	Female	55	2000	6
7	Female	54	1400	197
8	Female	56	1600	52
9	Female	60	1500	14
10	Male	60	1500	0
Mean		60.3	1770	86
Standard deviation (S.D)		9.70	309.30	

The high prevalence of *Contracaecum* spp infection in catfish may have implications for health of the fish and also for the potential definitive and accidental hosts that eat infected fish. In the present study, most of the third stage larval worms were found in the abdominal cavity, followed by the serosal surface of the intestine but not in the lumen or muscles. Most likely these worms could be discarded during evisceration of the fish. Although some workers (Mashego and

Saayman, 1981; Boomker, 1982; Paperna, 1996), did not report deleterious effects of the larvae *Contracaecum* spp on the health of the catfish, Tilapia species heavily infected with larvae of *Contracaecum* spp were found to be stunted with reduced fat deposits around the digestive caecum (Aloo, 2002); Acha and Szyfres 1987). In this investigation no larval worms were seen in the intestinal lumen but were abundant on the serosal surface of the intestine. Reports by Lymberry *et al.* (2002) indicate that the liver was the most parasitized organ. The heavy infection rates may partly be due to the fact that Clariid species are opportunistic feeders.

There was no evidence of post-mortem migration of the larva parasites from the abdominal cavity to musculature considering that the catfish was purchased long after their collection from the dam. Some authors have suggested that as soon as the host dies, the parasites migrate to the muscles (Paperna, 1996). This was not the case in this study.

Human anisakiasis occurs during accidental consumption of raw or undercooked fish. If eaten raw or undercooked, fresh fillets containing any anisakid roundworm may be considered a health hazard. Anisakid larval nematodes have been associated with abdominal pain, nausea and vomiting in acute stages. While chronic infection is associated with eosinophilic granulomatous reaction similar to Crohn's disease (Ito *et al.*, 2007). Although less invasive than other members of Anisakidae, larval of *Contracaecum* species have been shown to infect mammals (Vidal-Martinez *et al.*, 1994). Several cases of human infection with Anisakidae larvae nematodes have been documented (Noh *et al.*; 2003; Lopez-Serrano *et al.*, 2000; Muraoka *et al.*, 1996). Intestinal anisakidosis (Takei and Powell, 2007; Ito *et al.* 2007) and clinic gastric anisakidosis (Ito *et al.*, 2007) with Anisakid nematodes have been documented.

The finding of *Contracaecum* spp. in visceral organs of fish as predilection sites limits their zoonotic potential. Wharton *et al.*, (1999) suggested that if the fish are not frozen or filleted soon after capture the larval nematodes migrate into flesh. Although this

phenomenon was not observed in this study the demonstration of numerous worms in the majority of catfish examined should serve as a public health precaution.

## CONCLUSION

## REFERENCES

Acha, P.N. and Szyfres B (1987). Zoonoses and communicable diseases common to man and animals, 2<sup>nd</sup> ed Pan American Health Organization, Washington DC.

Aloo, P.A. (2001). Occurrence of larval Contracaecum (Nematoda: Heterocheillidae) in three teleostean species from Lake Naivasha, Kenya. *East African Journal of Science*, 3:1-12.

Aloo, P.A. (2002). A comparative study of helminth parasites from the fish *Tilapia zillii* and *Oreochromis leucostictus* in Lake Naivasha and Oloidien Bay, Kenya *Journal of helminthology*, 76: 95 – 104

Anderson, C. (1992). Nematode parasites of vertebrates. Their development and transmission. Commonwealth Agricultural Beurew, Wallingford, U.K. pp99

Barson, M. 2004. The occurrence of Contracaecum spp. Larvae (Nematoda: Anisakidae) in the catfish *Clarius gariepinus* (Burchell) from Lake Chivero, Zimbabwe. *Ondestepoort Journal of Veterinary Association*, 75:74-78.

Barson, M. and Avenant-Oldewage, A. (2006). Nematode parasites of *Clarius gariepinus* (Burchell, 1822) from the Rietvlei Dam, South Africa. *Ondestepoort Journal of Veterinary Association*, 73:87-94.

Boomker, J. (1982). Parasites of South African freshwater fish. I. Nematodes of the catfish [*Clarius gariepinus* (Burchell, 1822)] from the Hartbeespoort Dam. *Ondestepoort Journal of Veterinary Research*, 49: 41-51.

Cannon, L.R.G., (1977). Some larval ascaridoids from south-eastern Queensland marine fishes. *International Journal of Parasitology* 7:233-243.

Chabaud, A.G. (1974). Keys to subclasses, orders and super families, in CIH Keys to the nematode parasites of vertebrates, edited

To prevent human infections with anisakidac worms it is recommended that the catfish should be thoroughly cooked instead of being undercooked. Eating of raw fish is not allowed and catfish should be frozen to reduce the number of viable anisakidac worms

by R.C. Anderson, A.G. Chabaud & S. Wilmott. Slough: *Commonwealth Agricultural Bureaux*, 1:6-17.

Chishawa A.M.M. (1991). A survey of parasites of three *Siluriformes* [sic] fish species in Lake Kariba. University of Zimbabwe. University Lake Kariba Research Station Bulletin, 1/91.

Douëllou, L. (1992). A survey of fish parasites in Lake Kariba. University of Zimbabwe (University Lake Kariba Research Station Bulletin, 1/92).

Gomez, K.A. and Gomez, A.A. (1984) *Statistical Procedures for Agricultural Research*. 2<sup>nd</sup> edition Wiley, New York, USA pp367-371.

Hartwich, G. (1974). Keys to genera of the Ascaridoidea, in CIH Keys to the Nematode parasites of vertebrates, edited by R.C. Anderson and A.G. Chabaud.

Ito, Y., Ikemaysu, Y Yuzawa, H., Nishiwaki, Y., Kidah, H., Waki, S. Uchimura, M., Ozawa, T. Iwaoka, T. and Kanematsu, T. (2007). Chronic gastric anisakiasis presenting as pneumoperitoneum. *Asian Journal of Surgery* 30: 67 – 71.

Khalil, L.F. (1991). Techniques for identification and investigative helminthology: Techniques for processing platyhelminths and aconthophalans. In *Helminthology Manual*, edited by L.F. Khalil, International Institute of Parasitology, London, U.K.

Koie, M. and Fagerholm, H. (1995). The life cycle of *Contracaecum osculatum* (Rudolph, 1802) sensu stricto (Nematoda, Ascaridoidea, Anisakidae) in view of experimental infections. *Parasitological Research* 81:481-489.

Lopez-Serrano, M.C., Gomez, A.A. and Daschner, A. (2000). Gastroallergic anisakiasis: Finding in 22 patients. *Journal*

- of Gastroenterology and Hepatology*, 15: 503 - 506.
- Lymbery, A.J., Doupeç., Munshi, M.A. and Wong, T. (2002) Larvae of *Contracaecum* species among inshore fish species of southwestern Australia. *Diseases of Aquatic Organisms* , 51: 157- 159
- Malvestuto, S.P. and Ogambo-Ongoma, A. (1978). Observations of the infection of *Tilapia leucosticta* (Pisces: Cichlidae) with *Contracaecum* (Nematoda: Heterocheillidae) in Lake Naivasha, Kenya. *Journal of Parasitology*, 64:383-384.
- Mashego, S.N. and Saayman, J.E. (1981). Observations on the prevalence of the nematode parasites of the catfish *Clarius gariepinus* (Burchell, 1822), in Lebowa, South Africa. *South African Journal of Wildlife Research*, 11:46-48.
- Muraoka, A., Suchiro, I., Fujii, M. (1996). Acute gastric anisakiasis caused by Anisakiasis : 28 cases during the last 10 years. *Digestive Diseases Science* 41: 2362 - 2365.
- Noh, J.H., Kim B.J., Kim S.M., Ock, M.S., Park, M.I and Goo, J.Y. (2003) A case of acute gastric anisakiasis provoking severe clinical problems by multiple infection. Department of Parasitology, Department of Internal Medicine, Kosin Medical College Busan 602-703, Korea. Vol 41, No, 2, 97-100.
- Olson, A.C., Lewis, M.D. and Hauser, M.L (1983). Proper identification of anisakine worms. *American Journal of Medical Technology* : 49: 111 -114
- Paperna, I. (1996). Parasites, infections and diseases of fishes in Africa: An update FAO / CIFA Technical Paper, no.31.
- Takei, H., Powell, S.Z. (2007). Intestinal anisakidosis. *Annals of Diagnostic Pathology* : 12,20.
- Vidal-Martinez, V.M., Osorio-Sarabia, D. and Overstreet, R.M. (1994). Experimental infection of *Contracaecum multipapillatum* (Nematoda: Anisakidae) from Mexico in the domestic cat. *Journal of Parasitology* 80:576-579.
- Wharton D.A., Hassall M.L. and Alders O. (1999). Anisakis (Nematoda:) in some New Zealand inshore fish. *New Zealand Journal Marine and Freshwater Research* 33:643-648.
- Whifield, A.K. and Hegg, J. (1977). On the life cycles of the cestode *Ptychobothrium belones* and nematodes of the genus *Contracaecum* from Lake St. Lucia, Zululand, *South African Journal of Science*, 73:121-122.
- Yamaguti, S. (1959). *Systema Helminthum*. Vol. II. The cestodes of vertebrates. Interscience Publishers Inc vii, 860.
- Yamaguti, S. (1991) *Systema Helminthum*. Vol. III. The nematodes of vertebrates. New York & London: Interscience Publishers, Inc.1261