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Research Note

The first report of three acanthocephalan parasite species isolated from Philippine fishes

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Article info

Summary

Received January 6, 2015 Accepted August 28, 2015 Three acanthocephalan parasites, namely *Neoechinorhynchus* (*Neoechinorhynchus*) *quinghaiensis*, *Rhadinorhynchus ganapatti*, and *Bolbosoma* sp. are reported for the first time from Philippine fishes. *N. (N.) quinghaiensis* (Neoechinorhynchidae) may have been introduced into the country through the importation of carp species from China, where this parasite was first described and is presumed to be naturally occurring. The adult worms of *R. ganapatti* and the isolated cystacanth of *Bolbosoma* sp. represent the first record of parasites from both the family Rhadinorhynchidae and Polymorphidae in Philippine waters, respectively. These three new records encompass a third of all listed acanthocephalan parasites that have been reported in Philippine fish species to date, highlighting the need for more biodiversity-focused parasitology research, in light of potentially numerous more undocumented parasite species.

Keywords: Neoechinorhynchus; Bolbosoma; Rhadinorhynchus; fish parasites; Acanthocephala

Introduction

Parasites from the Phylum Acanthocephala encompass worms with a variety of recorded sizes, and a multitude of intermediate, paratenic, and final hosts (Amin, 1987). In the Philippines, the first records of acanthocephalan parasites were among fish species (Tubangui, 1933). Herein we report three new records of Acanthocephala isolated from Philippine fishes together with brief morphological descriptions, also coupled with a list of all recorded Acanthocephala associated with Philippine fishes as a reference.

Materials and Methods

Acanthocephalans were isolated from various fish species taken from field investigations last September to November 2013 in water bodies found South of Luzon Island, Philippines. All fish samples were dissected within 24 hours after capture. Isolated acanthocephalan specimens were first extended in cold water for 1 to 3 hours. These were then bound in slides under a cover slip together with pieces of thread for structural support, and were fixed in AFA

(10 mL formaldehyde, 10 mL acetic acid, 200 mL 70 % ethanol) for 24 h. These specimens were then pre-stained in 4 % FeNH, (SO₄)₂-H₂O, stained in Heidenhain's Hematoxylin, and decolorized again in 4 % FeNH₄ (SO₄)₂-H₂O until light brown, following a 1 to 3 day immersion series for each step depending on the size of each worm. After which, specimens were dehydrated in a series of ethanol concentrations, cleared in xylene, and mounted in Canada balsam. Parasite illustrations were made through the use of a drawing tube mounted on a light compound microscope (Olympus CX21). Listed measurements represent means of morphometric values, while range measurements represented the highest and lowest value observed when variations from multiple measurements were present. All values have been expressed as millimeters (mm), unless otherwise stated. Species identification was accomplished using keys from Amin (1987, 2002) and Amin et al. (2011). Slide preparations of the acanthocephalan parasites recovered in the study has been deposited as reference specimens in the Parasite Section of the Zoological Reference Collection of the University of Santo Tomas, Manila, Philippines (UST-ZRC-P).

Results and Discussion

We currently report three new records of Acanthocephala from Philippine fishes. Namely, *Neoechinorhynchus quinghaiensis*, *Rhadinorhynchus ganapatti*, and *Bolbosoma* sp. These three new records encompass a third of all the known species of acanthocephalan parasites associated with Philippine fishes (Table 1).

<u>Neoechinorhynchus (Neoechinorhynchus) quinghaiensis Liu,</u> Yang et Yang 1981

Description (3 adult males, 3 adult females): Trunk (Fig. 1a and b) aspinose, 3.5 long by 1.0 wide, four to five large hypodermic nuclei present. No visible neck, but with an evident restriction in anterior portion of trunk immediately before proboscis. Proboscis (Fig. 1c) small and globular, 0.12 long by 0.9 wide. Proboscis hooks arranged in three circles of six hooks each, with uniform hook size within each circle. Anterior hook circle largest, 50 μm long. Middle and posterior hook circles similar in size, 18 – 22 μm long. Proboscis receptacle short and single-walled. Leminisci elongate, subequal, extending beyond proboscis receptacle, 0.75 long by 0.5 wide. Testes post-equitorial. Anterior and posterior testis oblong, similar in size, 0.25 long by 0.20 wide. Cement glands syncytial. Everted copulatory bursa in males 0.35 long by 0.35 wide. Female reproductive system simple, 0.25 long. Unripe eggs (Fig. 1d) ellipsoidal with concentric membranes, 23 μm long by 11 μm wide.

We retrieved 24 specimens of Neoechinorhynchus (Neoechinorhynchus) quinghaiensis from Oreochromis niloticus (n=5), Hypophthalmichthys nobilis (n=2), and Carassius gibelio (n=1) from Lake Sampaloc and Laguna de Bay, November 22 and 27, 2013. De la Cruz & Paller (2012) previously reported the presence of Neoechinorhynchus sp. from O. niloticus in Lake Sampaloc. Their length measurements of proboscis hooks are slightly smaller (anterior proboscis hooks: 36.5±4.6 µm, middle and posterior proboscis hooks: 10.95±1.9 µm) than that reported in this study, but these seem to have been recorded from slightly smaller specimens (~2.0 mm inferred from Fig. 1 of de la Cruz & Paller (2012)). In addition, their qualitative descriptions of the shape and relative size of the trunk, proboscis, proboscis receptacle, and male and female reproductive organs fit our measurements of N. (N) quinghaiensis in the present study. The only difference in their report is the presence of "large and numerous cement glands", but this can be confidently argued as synctval based on their presented micrographs (Fig. 5 of de la Cruz & Paller (2012)). With these in mind, we conclude that previously reported Neoechinorhyncus sp. from Lake Sampaloc is indeed N. (N.) quinghaiensis. This is the first report of N. (N.) quinghaiensis occurring outside its original reported distribution from various cyprinid species in Qinhai Lake, China (Liqing et al., 1981; Tingbao & Xianghua, 2001), suggesting that this parasite may have been introduced into the country through the import of Chinese carp species.

Table 1. Provisional list of all acanthocephalan species that have been documented from fish in Philippine waters, including new records in the present study

Classification	First report	All recorded hosts	Locality
CAVISOMIDAE			
Cavisoma magnus	Regidor & Arthur, 1990	Chanos chanos	Mindanao
Filisoma rizalinum	Tubangui & Masiluñgan, 1946	Scatophagus argus	Luzon
DIPLOSENTIDAE			
Diplosentis amphacanthi	Tubangui & Masiluñgan, 1937	Siganus canaliculatus	Mindanao
ECHINORHYNCHIDAE			
Acanthocephalus sp.	Velasquez, 1976	Chanos chanos	Luzon, Mindoro
		Clarias macrocephalus	Laguna de Bay
NEOECHINORHYNCHIDAE			
Neoechinorhynchus sp.	de la Cruz & Paller, 2012	Oreochromis niloticus	Lake Sampaloc
Neoechinorhynchus octonucleatus	Tubangui, 1933	Leiopotherapon plumbeus	Luzon
Neoechinorhynchus quinghaiensis	this paper	Carassius gibelio	Lake Sampaloc
		Hypophthalmichthys nobilis	Laguna de Bay
		Oreochromis niloticus	Lake Sampaloc, Laguna de Bay
POLYMORPHIDAE			
Bolbosoma sp. (cystacanth)	this paper	Oreochromis niloticus	Lake Taal
RHADINORHYNCHIDAE Rhadinorhynchus ganapatti	this paper	Katsuwonis pelamis	Batangas Sea

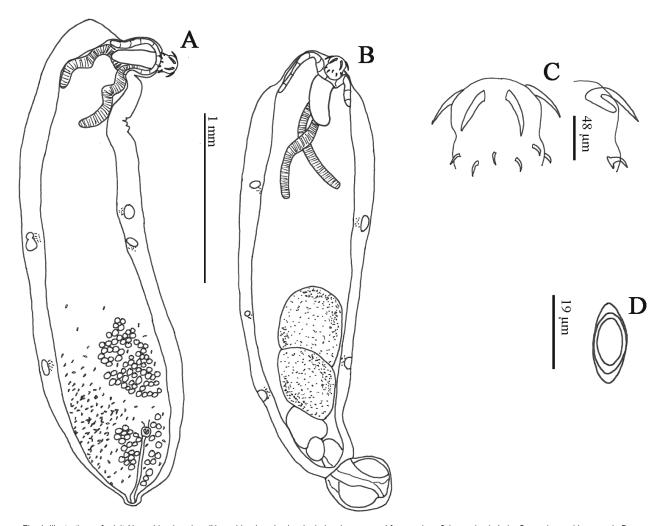


Fig. 1. Illustrations of adult Neoechinorhynchus (Neoechinorhynchus) quinghaiensis recovered from various fish species in Lake Sampaloc and Laguna de Bay, situated south of Luzon Island, Philippines. A: adult female, B: adult male, C: proboscis and proboscis hooks, D: egg

Rhadinorhynchus ganapatti Chandra, Rao et Shyamasunderi 1985

Description (2 adult males): Trunk (Fig. 2a) spinose, 6.25 - 6.38 (6.31) long by 0.68 wide, uniformly cylindrical. Trunk spines (Fig. 2b) present in anterior most region, in 5 circles of 5-8 spines per circle, 45 - 50 (48) long. Proboscis 1.38 long by 0.18 wide, uniformly cylindrical, with 16 longitudinal rows of 24 hooks each. Proboscis hooks (Fig. 2c) 48 – 58 (52) µm long, similar in shape and in their posteriorly directed angle of projection from proboscis, but are slightly tapering in length and width posteriorly. Hook roots simple, posteriorly directed. Basal hooks project more laterally as compared to anterior hooks. Proboscis receptacle 2.0 long by 0.18 wide. Leminsci equal, 1.33 long by 0.15 wide, does not extend past the proboscis receptacle, interspersed with small nuclei. Testes oblong, located in middle to anterior portion of trunk. Anterior testis 1.0 long by 0.45 wide, similar in size to posterior testis which is 0.95 long by 0.48 wide. Cement glands located immediately posterior to testes, 4 lobes in 2 pairs, which together are 1.42 long by 0.5 wide. Bursa 0.39 long by 0.5 wide.

We retrieved Rhadinorhynchus ganapatti from Katsuwonus pelamis (n=1) purchased from Lipa City fish market, October 1, 2013,

reportedly coming from fish catches taken from Batangas Sea. Two specimens of *R. ganapatti* were isolated from the liver and gills of the fish, but this is presumed to be the result of post mortem migration. *R. ganapatti* was originally described from marine fish species from India (see Amin *et al.*, 2011). The presence of *R. ganapatti* in the Philippines may be inferred from the cosmopolitan range of pelagic *K. pelamis* (Collette *et al.*, 1983). Our account of this parasite is the first report of an acanthocephalan species from the family Rhadinorhynchidae in the archipelagic waters of the Philippines, to date. The most geographically proximate report of Rhadinorhynchidae would be that of *R. trachuri* from benthic fish in the Pacific Ocean (Kovalenko, 1981).

Bolbosoma sp.

Description (1 female cystacanth): Trunk (Fig. 3a) spinose, 4.2 long by 1.09 wide in equatorial region with a tapering posterior region 0.4 wide, bulbous area present in trunk anterior 0.95 wide. Trunk spines (Fig. 3b) present in two distinct areas: anterior most region immediately before neck and in bulbous area, with the two spinose regions clearly demarcated by an aspinose zone. Anterior trunk spines arranged in 10 circles of 40 to 45 spines per circle, with each spine 0.13 long. Bulbous area trunk spines slightly

48 µm

Fig. 2. Illustration of adult *Rhadinorhynchus ganapatti* recovered from *Katsuwonus pelamis* reportedly caught from Batangas Sea, situated south of Luzon Island, Philippines. A: adult male, B: trunk spines, C: proboscis hooks

longer, 0.2 long, arranged in 8 circles with 60-66 spines per circle. Proboscis slightly everted, 0.95 long by 0.35 wide. Proboscis receptacle double-walled, 1.33 long by 0.4 wide. Leminsci subequal, 0.8-0.98 long by 0.19 wide, supported by mesentery that is attached throughout the trunk and twists in configuration immediately below the proboscis resceptacle. Neck short, 0.18 long by 0.4 wide. Proboscis hooks (Fig. 3c) arranged in 20 longitudinal

rows of 10 hooks per row, each hook 53 μm long. Female reproductive system simple, 88 μm long. Gonopore sub-terminal.

We retrieved one specimen of *Bolbosoma* sp. cystacanth encysted in the visceral wall of *Oreochromis niloticus* sampled from Lake Taal, September 11, 2013. We identified the cystacanth as Polymorphidae because of the presence of a double-walled proboscis receptacle and a spinose trunk (Amin, 1987). In addition, we identified the specimen based on the adult features of *Bolbosoma*, which has a characteristic bulbous anterior trunk, the presence

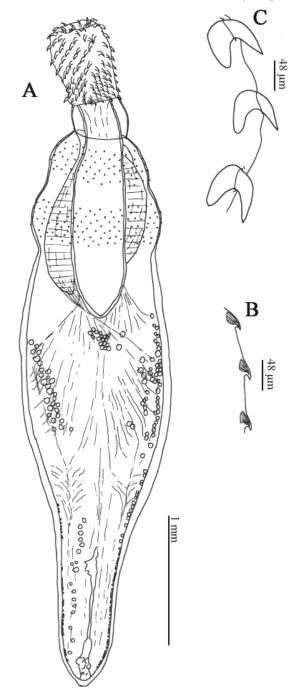


Fig. 3. Illustration of Bolbosoma sp. cystacanth recovered from Oreochromis niloticus in Lake Taal, situated south of Luzon Island, Philippines.
A: female cystacanth, B: trunk spines, C: proboscis hooks.

of two bands of trunk spines, and the absence of genital spines (Yamaguti, 1963). Polymorphidae cystacanths have visibly comparable features with adults in terms of proboscis armature and trunk spine arrangement (see Amin et al., 1995, 2010). Also, Bolbosoma can be differentiated from Corynosoma, which extends its trunk spines in the entirety of one side of the trunk and the presence of genital spines (e.g. Braicovich et al., 2005; Sardella et al., 2005; Mašová & Baruš, 2013). Our account of this parasite is the first report of an acanthocephalan species from the family Polymorphidae in the Philippines, to date. The most geographically proximate report of Polymorphidae would be that of Bolbosoma vasculosum and B. heteracanthe from marine pelagic and benthic fish in the Pacific Ocean (Kovalenko, 1981). In addition, this is also one of the very few reports of this cystacanth life stage to be retrieved from inland water bodies, since adult stages of the genera Bolbosoma utilize marine mammals as final hosts (see Santos et al., 2008). This may be attributed to the unique limnological features of Lake Taal (Papa & Mamaril, 2011) with its slightly elevated salinity (among other unique characteristics) that has allowed the remarkable freshwater speciation of previously landlocked marine species (e.g. Sardinella tawilis, Hydrophis semperi). The presence of Bolbosoma sp. cystacanth in the lake may suggest the continued migratory activity of various fish species, which may be utilized as paratenic hosts, from Lake Taal going into and from Batangas Sea through Pansipit River.

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References

AMIN, O. M. (1987): Key to the families and sub-families of Acanthocephala, with the erection of a new class (Polyacanthocephala) and a new order (Polyacanthorhynchida). *J. Parasitol.*, 73(6): 1216 – 1219. DOI: 10.2307/3282307

AMIN, O. M. (2002): Revision of *Neoechinorhynchus* Stiles & Hassall, 1905 (Acanthocephala: Neoechinorhynchidae) with keys to

88 species in two subgenera. *Syst. Parasitol.*, 53: 1 – 18. DOI: 10.1023/A:1019953421835

AMIN, O. M., HECKMANN, R. A., MESA, R., MESA, E. (1995): Description and host relationships of cystacanths of *Polymorphus spindlatus* (Acanthocephala: Polymorphidae) from their paratenic hosts in Peru. *J. Helminthol. Soc. Wash.*, 62(2): 249 – 253

AMIN, O. M., HECKMANN, R. A., PEÑA, C., CASTRO, T. (2010): On the larval stages of *Polymorphus spindlatus* (Acanthocephala: Polymorphidae) from a new fish host, *Oreochromis niloticus*, in Peru. *Neotrop. Helminthol.*, 4(1): 81 – 85

AMIN, O. M., HECKMANN, R. A., NGUYEN, V. H. (2011): Description of two new species of *Rhadinorhynchus* (Acanthocephala, Rhadinorhynchidae) from marine fish in Halong, Bay Vietnam, with a key to the species. *Acta Parasitol.*, 56(1): 67 – 77. DOI: 10.2478/s11686-011-0004-3

Braicovich, P., Gonzalez, R. A., Tanzola, R. D. (2005): First record of *Corynosoma australe* (Acanthocephala, Polymorphidae) parasitizing seahorse, *Hippocampus* sp. (Pisces, Syngnathidae) in Patagonia (Argentina). *Acta Parasitol.*, 50(2): 145 – 149

COLLETTE, B. B., NAUEN, C. E. (1983): Scombrids of the world. An annotated and illustrated catalogue of tunas, mackerels, bonitos and other related species known to date, FAO species catalogue Vol. 2. Rome, Food and Agriculture Organization of the United Nations, 137 pp.

DE LA CRUZ, C. P. P., PALLER, V. G. V. (2012): Occurence of *Neoechinorhynchus* sp. (Acanthocephala: Neoechinorhynchidae) in cultured Tilapia, [*Oreochromis niloticus* (L.), Perciformes: Ciclidae] from Sampaloc Lake Philippines. *Asia Life Sci.*, 21(1): 287 – 298. KOVALENKO, L. M. (1981): Acanthocephalans of some fish in the tropical zone of the Pacific Ocean. *Zool. Zhurnal* 60(8): 1140 – 1144

LIQING, L., BAODUE, W., TAO, Y. (1981): Notes on the parasites of the naked carp of Qinghai Hu. *J. Fish. China*, 5(4): 295 – 299

Mašová, S., Baruš, V. (2013): Redescription of cystacanths of *Corynosoma pseudohamanni* Zdzitowiecki, 1984 (Acanthocephala: Polymorphidae) from paratenic fish hosts. *Folia Parasitol.*, 60(2): 169 – 176. DOI: 10.14411/fp.2013.019

Papa, R. D. S., Mamaril, Jr., A. C. (2011): History of the biodiversity and limno-ecological studies on Lake Taal with notes on the current state of Philippine limnology. *Philipp. Sci. Lett.*, 4(1): 1 – 10. Regidor, S., Arthur, J. R. (1990): A survey of the parasite of milkfish (*Chanos chanos*) in the Philippines. In *Abstracts of Symposium on Diseases in Asian Aquaculture by the Fish Health Sector of the Asian Fisheries Society, November 26 – 29, 1990.* Bali, Indonesia, pp. 34

Santos, C. P., Gibson, D. I., Tavarez, L. E., Luque, J. L. (2008): Checklist of Acanthocephala associated with the fishes of Brazil. Zootaxa, 1938: 1-22

SARDELLA, N. H., MATTIUCCI, S., TIMI, J. T., BASTIDA, R. O., RODRIGUEZ, D. H., NASCETTI, G. (2005): *Corynosoma australe* Johnston, 1937 and *C. cetaceum* Johnston & Best, 1942 (Acanthocephala: Polymorphidae) from marine mammals and fishes in Argentinian waters: allozyme markers and taxonomic status. *Syst. Parasitol.*, 61: 143 – 156. DOI: 10.1007/s11230-005-3131-0

TINGBAO, Y., XIANGHUA, L. (2001): Seasonal population dynamics of *Neoechinorhynchus qinghaiensis* in the carp, *Gymnocypris prze-*

walskii przewalskii, from Qinghai Lake, China. *J. Helminthol.*, 75: 93 – 98. DOI: 10.1079/JOH200030

Tubangui, M. A. (1933): Notes on Acanthocephala in the Philippines. *Philipp. J. Sci.*, 50: 115 – 128

Tubangui, M. A., Masilungan, V. A. (1937): *Diplosentis amphacanthi* gen. et sp. nov., an Acanthocephala parasitic in marine fish. *Philipp. J. Sci.*, 62: 83 – 189

Tubangui, M. A., Masiluñgan, V. A. (1946): On two acanthocephala from the Philippines. $J.\ Parasitol.,\ 32:\ 154-155$

Velasquez, C. C. (1976): Fish parasitology in conservation and public health. *Fish. Res. J. Philipp.*, 1: 52 – 55

YAMAGUTI, S. (1963): Systema Helminthum: Acanthocephala. 5th Volume, John Wiley & Sons, Inc., 423 pp.