LIGHT AND SCANNING ELECTRON MICROSCOPIC STUDY OF *RHABDOCHONA BILQEESAE* SP. N. (NEMATODA: RHABDOCHONIDAE) PARASITIZING A COMMON EDIBLE FISH *LABEO ROHITA* IN THATTA, SINDH, PAKISTAN

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ABSTRACT

Present studies under Light and Scanning Electron Microscopic observations of a spiruroid nematode *Rhabdochana bilqeesae* sp. n. found parasitizing a common edible fresh water fish *Labeo rohita* in district Thatta, Sindh, Pakistan. Species of the genus are most frequently recorded worldwide including Pakistan and seems to be bound to the fresh waters only.

The new species *Rhabdochona bilqeesae* is characterized by having well developed cephalic and cervical alae in both male and female specimens; the number of teeth in the prostom appears to be ten. The coiled caudal region in the male specimens are provided with spiny patches some distance above the spicules; The tail tip in the males is roughly conical and some distance above the tip, there are 4-5 rows of scale like structures which have not been reported earlier. The tail tip in females is roughly rounded. The eggs are embryonated and have smooth walls. *Labeo rohita* is a new host record of this nematode in Pakistan.

Key words: Rhabdochona bilqeesae sp.n. Labeo rohita, Dist. Thatta, Sindh, Pakistan

INTRODUCTION

Species of the genus *Rhabdochona Railliet*, 1916 are widely distributed worldwide. Nineteen Species of the genus have been recorded from fresh water fishes in Pakistan.

Present studies are based upon light and scanning electron microscopic observations of *Rhabdochona bilqeesae* sp.n. recorded during an investigation on prevalence of helminth parasitic infections in Sindh, Pakistan.

Present specimens however appear different in having pseudolabia provided with a pair of labial papillae; well developed cephalic and cervical alae; absence of deirids; caudal region in male specimens have spiny patches and caudal end in having few rows of prominent scale like structures situated a little above the tail-tip. The eggs are embryonated with smooth walls, and *Labeo rohita* is a new host record in Sindh, Pakistan.

Rhabdochona species reported from Pakistan are: Rhabdochona magna (Khan and Yaseen, 1969) Zaidi and Khan, 1975 from Rita rita, R. cavasius Rehana and Bilqees, 1973 from Mystus cavasius; R. chanawensis Zaidi and Khan, 1975 from Eutroiichthys vacha; R. (Filochona) charsaddiensis Siddiqui and Khattak, 1984 from Puntius sp. and Nemachilus sp.; R. (F.) schizothoracis Siddiqui and Khattak, 1984 from Schizothorax habiatus and S. plagiostomus; R. megasacculata Ghazi et al., 1999 from Brilius vagra; R. sarana (Karve et Naik 1951) Akram and Khatoon, 2001; R. hellichi (Sramek, 1901) Akram and Khatoon, 2001 from Schizothorax plagiostomus; R. (Globochona) rahimi Ghazi et al., 2003 from Brilius pakistanicus; R. kharani Kakar et al, 2006; from Labeo gedrosicus; R. nushkiai Kakar and Bilqees, 2007 from Cyprinion milesi and C. watsoni; R. watsoniai Kakar and Bilqees 2007, from the fish Cyprinion watsoni; R. uvaginus Kakar and Bilqees, 2007 from Tor putitora; R. (R.) hingoli Kakar and Bilqees, 2007 from Cyprinion milesi and Rastrelliger kanagurta; R. bifidum Kakar and Bilqees, 2007 from Tor putitora; R. magnavesicula Kakar and Bilqees, 2008 from Schizocyprus brucei; R.(R.) milesi Kakar et al., 2008 form Cyprinion milesi; R.(R.)mujibi Kakar and Bilqees, 2009 from Tor putitora; R.(F.) gubernaculus Kakar et al., 2010 from Cyrinion watsoni.

MATERIALS AND METHODS

Labeo rohita is an economically important edible fish in District Thatta, Sindh. More than a dozen fish were caught from Thatta and brought to the Laboratory for examining parasitic infections. Three male and nineteen female nematodes were recovered from the small intestine of a fish. The nematodes were first studied alive in

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normal saline under a binocular, later these were fixed in hot, steaming alcohol and stored in alcohol glycerin mixture (1 part glycerin: 9 parts 70% ethanol). For detailed study the nematodes were cleared in glycerin. The cephalic, cervical and caudal region of male specimens were also examined with Scanning Electron Microscope. Light photomicrographs were prepared with the curtsey of Department of Zoology, University of Karachi while Scanning Electron Micrographs were prepared at Central Science Laboratory, University of Karachi. Eggs were dissected out from the uterus of a damaged female. All the drawings were made with the aid of a camera Lucida. The specimens are deposited at Aquaculture and Fisheries Program, Animal Sciences Institute, National Agricultural Research Centre (NARC), Islamabad, Pakistan.

Rhbdochona bilqeesae sp.n. (Figs. 1-15)

Host: *Labeo rohita* (Ham.) Site of infection: Small intestine

Locality: Thatta district, Sindh, Pakistan Number of hosts examined / infected: 15/1 Number of specimens recovered 3%, 19

Percentage of infection: 6.6%

Intensity of infection: 1–21 (worms per fish)

Description is based upon 3 male and 19 female specimens.

These are thin, delicate, and medium sized nematodes. The cuticle is transversely striated. Pseudolabia are evident through S.E.M. provided with a pair of labial papillae. The cephalic and cervical regions are provided with well developed cephalic and cervical alae. Two fairly large, lateral amphids are evident. The prostom is thick-walled and funnel-shaped and bears on its inner surface chitinised ridges which roughly appear to be 10 in number. The prostom or the buccal capsule opens into a vestibule or mesostom, which is moderately long and leads to the anterior-muscular and posterior glandular oesophagus. The intestine is simple. Deirids are not conspicuous. Tail in male ends in a roughly conical end. The eggs are embryonated and have smooth walls.

Male

Body cuticle is finely striated transversely. Body length ranges from 10.4–15.6 (13.0) by 0.1–0.12 (0.11). The prostom is funnel or cup shaped 0.029-0.034 (0.031) by 0.036–0.039 (0.037), the mesostom is long and thin 0.15 – 0.17 (0.16) by 0.014–0.016 (0.015). The muscular oesophagus is also long and thin 2.2–2.5 (2.35) by 0.25–0.08 (0.069) while the glandular oesophagus is 3.4–4.0 (3.7) by 0.2–0.22 (0.21), leads to a simple intestine which opens through the rectum to the anal opening in the caudal region. The anterior end is provided with a well developed cephalic and cervical alae 2.0–2.2 (2.1) long and 0.02–0.034 (0.027) wide. In some specimens the cephalic alae extends up to smaller length (Figs. 3 & 12) while in others the alae extends up to the mesostom length (Figs. 1 & 4) and in some other specimens it extends up to the length of muscular oesophagus (Fig. 5). The mouth opening is guarded by labia or lips provided with a pair of papilla. The funnel shaped prostom is provided with longitudinal ridges which end in chitinous teeth like structures; these appear to be ten in number. Deirids, excretory pore and the nerve ring are not obvious in the specimens studied. Spicules are unequal and dissimilar. The left spicule is moderately

Long, while the smaller right spicule is bent inside and boat-shaped. The left spicule measure 1.20–1.22, and the right spicule is 0.42–0.43 long ending in a bluntly conical tip. Length ratio of the spicules is 1: 2.8–2.8 Tail 0.3–0.34 (0.32) long.

There are peculiar spiny patches on the inner lateral side of the coiled caudal region, which start some distance above the left spicule and continue forward up to some distance (Figs. 2, 12). There are 13 pairs of pedunculate preanal papillae and five pairs of post anal papillae, the fifth pair of the post anal papillae lie a little on the lateral side.

The cuticular striations are very prominent in the pre and post anal region. The tail tip is roughly conical and some distance above the tail-tip there are five rows of scale like structures, not reported earlier (Fig. 15).

Female

Description is based upon 19 mature egg bearing specimens:

Delicate, thin worms. Body cuticle striated finely throughout. Anterior posterior ends are tapering with posterior end more pointed, ending roughly into a rounded end. The most striking feature is the presence of cephalic and cervical alae, occupying different cephalic and cervical lengths and shapes Figs. 3, 4, 5 and 6. Oral aperture roughly

oval, with outwardly directed labia and a pair of labial papillae. Prostom funnel-shaped with interior margin armed with 10 small, forwardly directed teeth. Vestibule or mesostom moderately long. Deirids not conspicuous. Muscular oesophagus moderately long. Glandular esophagus more than twice the length of muscular oesophagus it occupies the whole width of the body below the muscular esophagus. Tail tip is rounded, taper sharply below the anal opening, ends into a rounded-tip. Body length 16.24–22.64 (19.44), maximum width in the post equatorial region 0.25–0.41 (0.33) Prostom 0.12–0.16 (0.13) long and 0.08–0.15 (0.11) wide in lateral view. Length of vestibule including prostom is 0.31–0.60 (0.48). Muscular oesophagus 0.82–1.4 (1.11) long, maximum width 0.08–0.09 (0.085); glandular esophagus comparatively longer and stout 1.45–1.90 (1.67) long, maximum width 0.125–0.13 (0.27); length ratio of both parts 1:1.3–1.35. Nerve ring, excretory pore and deirids are not conspicuous. Vulva post equatorial 8.19–12.06 (10.45) from the anterior extremity. Vulval flap forwardly directed with fairly long vaginal tube (Fig. 7) which is directed posteriorly from Vulva. Eggs oval, embryonated and have smooth walls, 0.071–0.074 (0.072) by 0.06–0.061. Tail 0.62–0.64 (0.63) long.

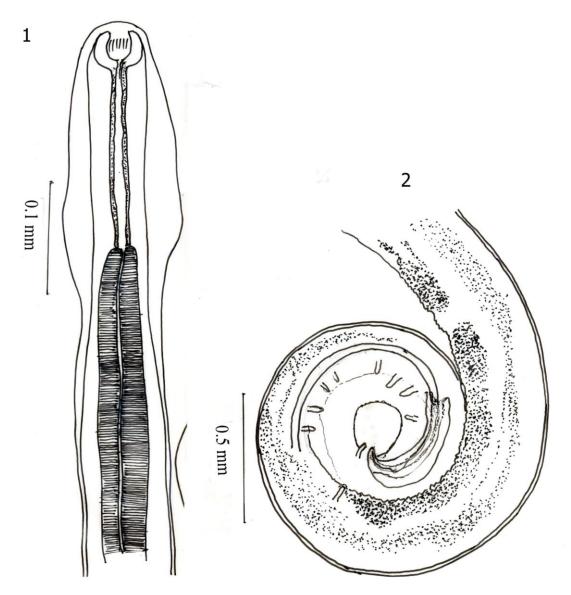


Fig.1.-2: Rhabdochona bilgeesae sp.n., holotype, male;

- 1: Anterior region, lateral view showing pro and mesostom, part of muscular oesopgagus and well developed cephalic and cervical alac.
- 2: Coiled caudal region, lateral view showing spicules, pedunculate caudal papillae and typical spiny patched.

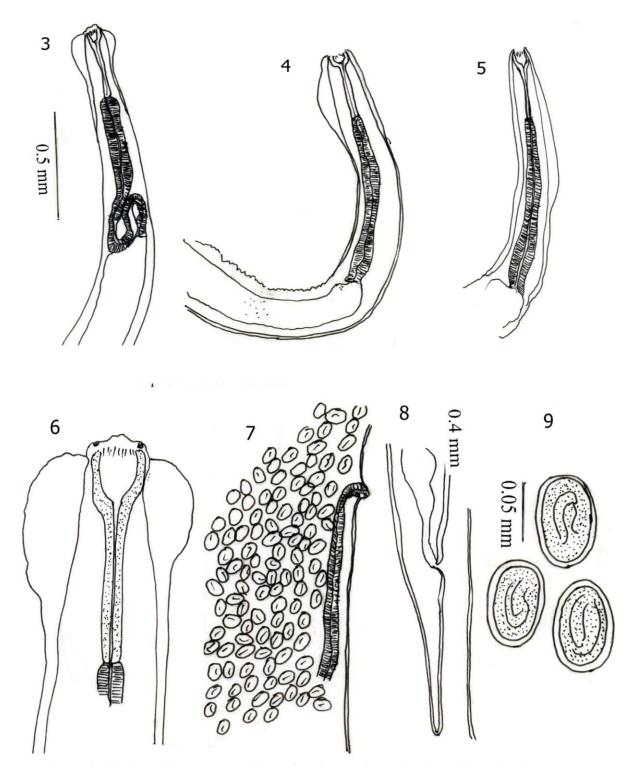


Fig.3-9. Rhabdochona bilqeesae sp.n. allotype, female;

- 3: Anterior region, lateral view showing smaller cephalic alae.
- 4-5: Anterior region lateral view paratype specimensshowing longer cephalic and cervical alae.
- 6: Anterior region lateral view showing smaller cephalic alae, pro and mesostom.
- 7: Region of vulva and vagina with uteus packed with eggs.
- 8: Caudal region showing tail with bluntly rounded end.
- 9: Embryonated eggs with smooth walls.



Fig.10-11. Rhabdochona bilqeesae sp.n. photomicrograph.

- 10: Male holotype, anterior region showing well developed cephalic and cervical alae, pro mesostome, muscular and part of grandular oesophagus. 10x.
- 11: Caudal region tight coiled, lateral view showing typical spiny patches above the laft spicule, right and left spicules and pedunculate caudal papillae. 5x20.
- Figs. 12-14: Rhabdochana bilgeesae sp.n. allotype female photomicrographs.
- Fig.12: Anterior region showing cephalic alae, pro and mesostom, muscular and part of grandular oesophagus.
- Fig.13: Vulva, vagina and part of filled with ova; Fig.14: Cauda; region lateral view showing anal opening tail and roughy rounded tail tip.



Fig.15. Rhabdochona bilqeesae sp.n. Paratype male scanning Electron micrograph. Caudal tip region, lateral view enlarged showing 4-5 rows od scales and bluntly pointed tail tip.

DISCUSSION

More than 100 species of the genus *Rhabdochona* Railliet, 1916 have been reported, almost all from fresh water fishes, from almost all zoogeographical regions (Moravec, 1975; Moravec *et al.*, 2012). Nineteen species of the genus have been reported from fresh water fishes in Pakistan.

Present species is described from a common edible fish Labeo robita caught from Thatta District in Sindh.

Some striking features such as: presence of cephalic and cervical alae which extends up to the length of prostom, mesostom and up to muscular esophagus in some specimens (Figs 1,3,4,5 & 6), presence of pseudolabia; absence of deirids; presence of minute spiny patches a little above the left spicule which continue forward up to some distance. Thin-shelled embryonated eggs with smooth walls, thirteen pairs of pre-anal papillae and five pairs of post-anal papillae including a lateral pair, and above all four-five rows of scale-like structures situated some distance above the caudal tip (Fig. 15) differentiate the present specimens from earlier reported species of Pakistan and elsewhere.

A new species *Rhabdochona bilqeesae* is therefore proposed. Species name refers to the Renowned Parasitologist Dr. Bilqees Fatima Mujib of Pakistan.

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