

Description of *Rhabdochona (Globochona) rasbora* sp. n. (Nematoda: Rhabdochonidae) from the freshwater cyprinid fish *Rasbora paviana* Tirant in southern Thailand

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Abstract: A new nematode species, *Rhabdochona (Globochona) rasbora* sp. n. (Rhabdochonidae), is described from the intestine of the freshwater cyprinid fish (sidestripe rasbora) *Rasbora paviana* Tirant in the Bangbaimai Subdistrict, Muang District, Surat Thani Province, southern Thailand. It differs from other representatives of the subgenus *Globochona* Moravec, 1972 which possess eggs provided with lateral swellings in having a spinose formation at the tail tip of both sexes and in some other morphological features, such as the body length of gravid female (8.6–23.7 mm), presence of two–three swellings on the egg, eight anterior prostomal teeth, length ratio of spicules (1 : 5.3–6.7) and arrangement of male genital papillae. This is the third nominal species of *Rhabdochona* Railliet, 1916 and the second species of the subgenus *Globochona* reported from fishes in Thailand. The three species of *Rhabdochona* recently described from fishes in Pakistan, viz. *R. annai* Kakar, Bilqees et Khan, 2012, *R. bifurcatum* [sic] Kakar et Bilqees, 2012, and *R. pakistanica* Kakar, Bilqees et Khan, 2012, are considered to be *species inquirendae*.

Keywords: parasitic nematode, *Rhabdochona*, *Globochona*, freshwater fish, sidestripe rasbora, Cyprinidae, Thailand

As recently stated by Moravec and Yooyen (2011a), the fauna of freshwater fish nematodes in Thailand remains little known. The records of these parasites in Thailand inland waters have been rather scarce (Pearse 1933, Ratanasritong and Kliks 1972, Sirikanchana 1982, Moravec and Scholz 1991, Moravec et al. 1999, 2004, 2006, 2007, 2011, Wongsawad et al. 2004, Boonchot and Wongsawad 2005, Purivirojkul 2009, Moravec and Yooyen 2011a,b) and the reported parasites often were not identified to species. Kamchoo (2000) [presently transcribed from Thai as Kamchoo] was the first to record representatives of the genus *Rhabdochona* Railliet, 1916 in her unpublished thesis, reporting four unidentified species, designated as *Rhabdochona* sp. and *Rhabdochona* spp. I–III, from nine species of fishes in the Maesa Stream, Doi Suthep-Pui National Park, Chiang Mai, northern Thailand. Later the same unidentified *Rhabdochona* spp. from fishes of this locality were reported in the paper of Wongsawad et al. (2004). Two new species of *Rhabdochona*, *R. (Rhabdochona) pseudomysti* Moravec et Yooyen, 2011 and *R. (Globochona) thaiensis* Moravec et Yooyen, 2011, have recently been described from *Pseudomystus siamensis* (Regan) and *Mystacoleucus marginatus* (Valenciennes), respectively, from northern Thailand (Moravec and Yooyen 2011a).

During recent helminthological investigations on some freshwater fishes in the Surat Thani Province, southern Thailand, nematodes referable to *Rhabdochona* were recovered from the intestine of the sidestripe rasbora *Rasbora paviana* Tirant (Cyprinidae, Cypriniformes). Their detailed study using the light and scanning electron microscopy showed that they represent a previously undescribed species belonging to the subgenus *Globochona* Moravec, 1972. This species is described herein.

Rasbora paviana (maximum body length 12 cm) is a tropical freshwater fish of minor commercial importance, which is native in Thailand. Its distribution includes the Mekong, Chao Phraya and Meklong basins, being also reported from the northern Malay Peninsula (Froese and Pauly 2012).

MATERIALS AND METHODS

Fish were collected by a local fisherman using gill nets from a small tributary creek of the Tapi River in the Bang Baimai Subdistrict, Muang District, Surat Thani Province. Thirty-five specimens of *Rasbora paviana* (total body length 9.0–12.8 [mean 10.3] cm, weight 4–18 [9.3] g) were examined for parasites during March and April of 2011. The nematodes recovered were washed in physiological saline and then fixed in cold 4% formaldehyde solution. For light microscopy (LM) examina-

tion, the nematodes were cleared with glycerine. Drawings were made with the aid of a Zeiss drawing attachment. Specimens used for scanning electron microscopy (SEM) were postfixed in 1% osmium tetroxide (in phosphate buffer), dehydrated through a graded acetone series, critical-point-dried and sputter-coated with gold; they were examined using a JEOL JSM-7401F scanning electron microscope at an accelerating voltage of 4 kV (GB low mode). All measurements are in micrometres unless otherwise indicated. The names of fishes follow FishBase (Froese and Pauly 2012).

RESULTS

Family Rhabdochonidae Travassos, Artigas et Pereira, 1928

Rhabdochona (Globochona) rasbora sp. n.

Figs. 1, 2

Description: Medium-sized nematodes with finely transversely striated cuticle. Oral aperture hexagonal to oval, with four submedian sublabia, surrounded by four small submedian cephalic papillae and pair of lateral amphids (Figs. 1E, 2A,B). Prostom barrel-shaped, with distinct basal teeth (Fig. 1C,D). Anterior margin of prostom armed internally with eight fairly large, forwardly directed teeth (two dorsal, two ventral and two on each side) (Figs. 1E, 2A,B). Vestibule long (Fig. 1A–C). Deirids small, bifurcate, situated at posterior half of vestibule (Figs. 1B,C,F, 2H). Glandular oesophagus 8–15-times longer than muscular oesophagus, occupying entire width of body (Fig. 1A). Tail of both sexes conical with truncated tip, optically forming two minute (dorsal and ventral) posteriorly oriented spikes (Fig. 1N,O); in fact, this is ring of elevated cuticle with sharp edge, bearing about three thickenings; small terminal mucron provided with many minute spine-like projections arises from middle of above-mentioned cuticular ring (Fig. 2F,G).

Male (seven specimens; measurements of holotype in parentheses): Length of body 5.55–6.12 (5.63) mm, maximum width 68–82 (68). Prostom 12 (12) long and 12 (12) wide in lateral view. Length of vestibule including prostom 114–120 (117). Muscular oesophagus 135–240 (168) long, maximum width 15–21 (15); glandular oesophagus 1.56–2.01 (1.77) mm long, maximum width 42–60 (48); length ratio of both parts 1:8–13 (1:11). Length of vestibule with prostom and entire oesophagus forms 33–40 (35)% of body length. Nerve ring encircling muscular oesophagus 156–174 (171) from anterior extremity; excretory pore at 210–246 (228). Deirids not located. Preanal papillae: 7–8 (7) subventral and 1 (1) lateral pairs; latter situated at about level of, or somewhat posterior to, third pair of subventrals (counting from cloacal opening) (Figs. 1S, 2D). Postanal papillae: 6 (6) pairs, of which 5 (5) pairs subventral and 1 (1) lateral; lateral pair approximately at level of first pair of subventrals or slightly posterior to it (Figs. 1S, 2C,D). Precloacal ventral cuticular ornamenta-

tion (area rugosa) formed by numerous transverse bands separated from each other by single transverse groove, each provided with band-like field bearing many low longitudinal, parallel ridges (Figs. 1K, 2E). Left spicule 399–456 (441) long; its shaft 75–108 (90) long, representing 16–26 (20)% of entire spicule length; distal tip of extruded spicule slightly widened, blunt (Figs. 1R,S, 2C,D). Right spicule boat-shaped, 66–81 (66) long, without dorsal barb at distal tip (Fig. 1P,S). Length ratio of spicules 1:5.3–6.7 (1:6.7). Tail 141–204 (204) long (Figs. 1S, 2C,D); terminal mucron bearing cuticular spines 6 (6) long and 4.5–6 (6) wide (Figs. 1N,S, 2C,D,F,G).

Female (five gravid specimens with mature eggs; measurements of allotype in parentheses. Measurements of five nongravid and those containing few immature eggs in square brackets): Length of body 8.64–23.72 (12.68) [4.27–6.64] mm, maximum width 109–272 (150) [68–95]. Prostom 15–21 (21) [9–18] long and 12–18 (18) [9–15] wide in lateral view. Length of vestibule including prostom 114–156 (132) [102–126]. Muscular oesophagus 165–198 (198) [153–165] long, maximum width 21–30 (30) [15–21]; glandular oesophagus 2.12–2.86 (2.86) [1.51–2.07] mm, maximum width 60–75 (75) [42–57]; length ratio of both parts 1:12–15 (1:14) [1:10–13]. Length of vestibule with prostom and entire oesophagus forms 12–28 (25) [29–41]% of body length. Nerve ring, excretory pore and deirids at 168–213 (213) [150–168], 204–294 (276) [210–243] and 57–117 (not located) [69–87], respectively, from anterior extremity. Vulva post-equatorial, 4.76–13.15 (7.48) [2.54–3.92] mm from anterior extremity, at 53–65 (59) [58–64]% of body length. Vagina directed first anteriorly and then posteriorly from vulva (Fig. 1L). Mature eggs oval, thick-walled, larvated, provided with two large oval, almost opposite lateral swellings (floats) (Fig. 1G,H); some eggs bearing one additional, smaller and almost circular equatorial swelling situated between two large swellings (Fig. 1I,J); swellings absent or poorly developed on immature eggs. Size of mature eggs 39–42 × 18–21 (39–42 × 18–21), egg-wall 3 (3) thick, swellings 6–9 (6–9) high. Tail 237–340 (246) [153–286] long (Fig. 1M); caudal mucron bearing spines 6 (6) [5–6] long and 5–6 (6) [6] wide.

Type host: Sidestripe rasbora, *Rasbora paviana* Tirant, 1885 (Cyprinidae, Cypriniformes).

Site of infection: Intestine.

Type locality: Small tributary creek of the Tapi River, Bang Baimai Subdistrict, Muang District, Surat Thani Province, southern Thailand (collected in March and April 2011).

Prevalence and intensity of infection: 40% (14 fish infected/35 fish examined); 1–7 (mean 3) nematodes per fish.

Deposition of type specimens: Helminthological Collection of the Institute of Parasitology, BC AS CR in České Budějovice, Czech Republic (Cat. No. N-994).

Etymology: The specific name *rasbora* (genitive) relates to the generic name of the host fish (*Rasbora*).

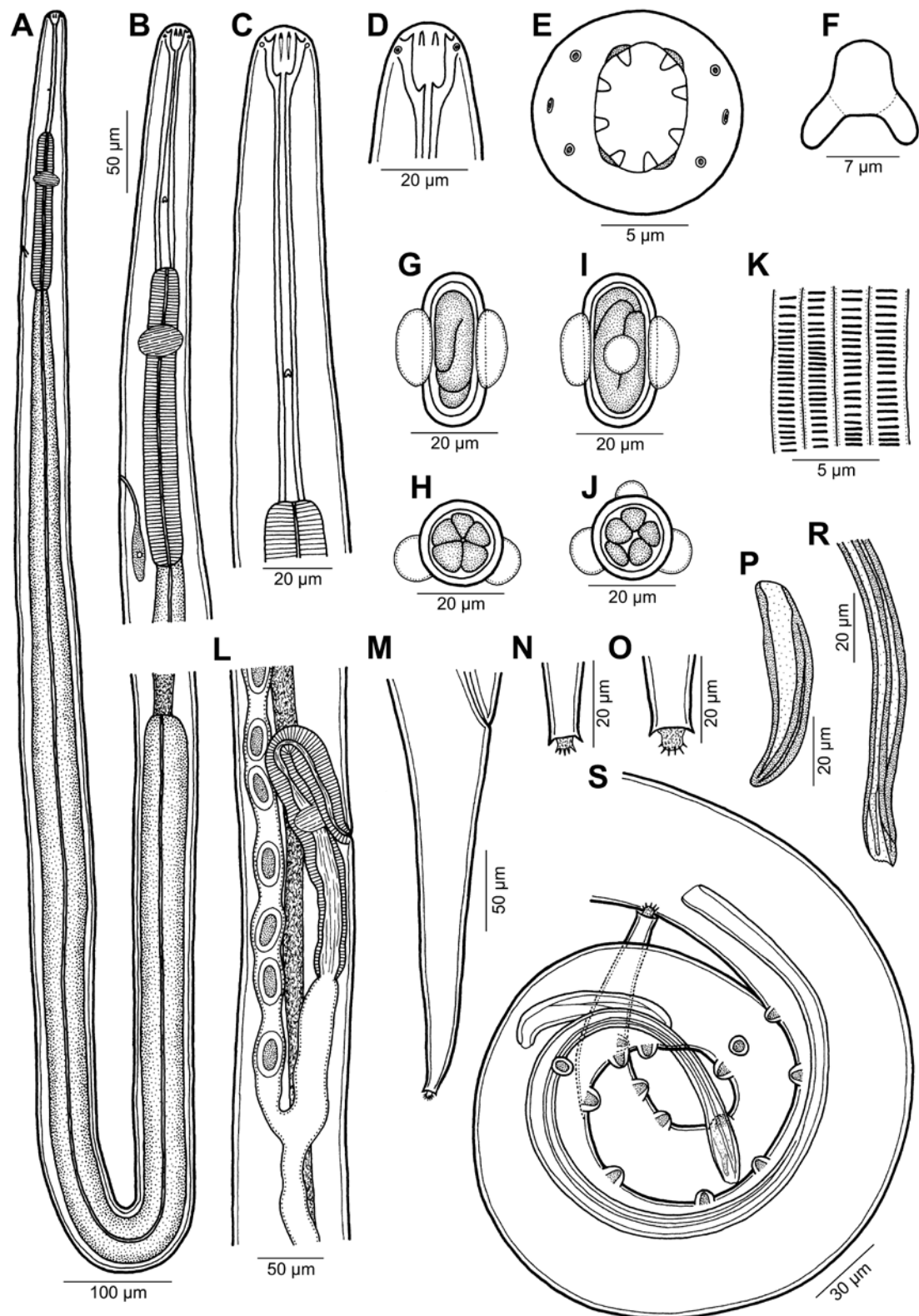


Fig. 1. *Rhabdochona (Globochona) rasborae* sp. n. **A** – oesophageal part of male, lateral view; **B** – anterior end of female, lateral view; **C** – region of vestibule of largest female, lateral view; **D**, **E** – cephalic end, lateral and apical views; **F** – deirid; **G**, **H** – egg with two swellings, lateral and apical views; **I**, **J** – egg with three swellings, lateral and apical views; **K** – detail of male ventral precloacal ornamentation; **L** – region of vulva of female containing immature eggs, lateral view; **M** – tail of female, lateral view; **N**, **O** – tail tip of male and female, respectively; **P** – right spicule, lateral view; **R** – distal end of left spicule, lateral view; **S** – posterior end of male, lateral view.

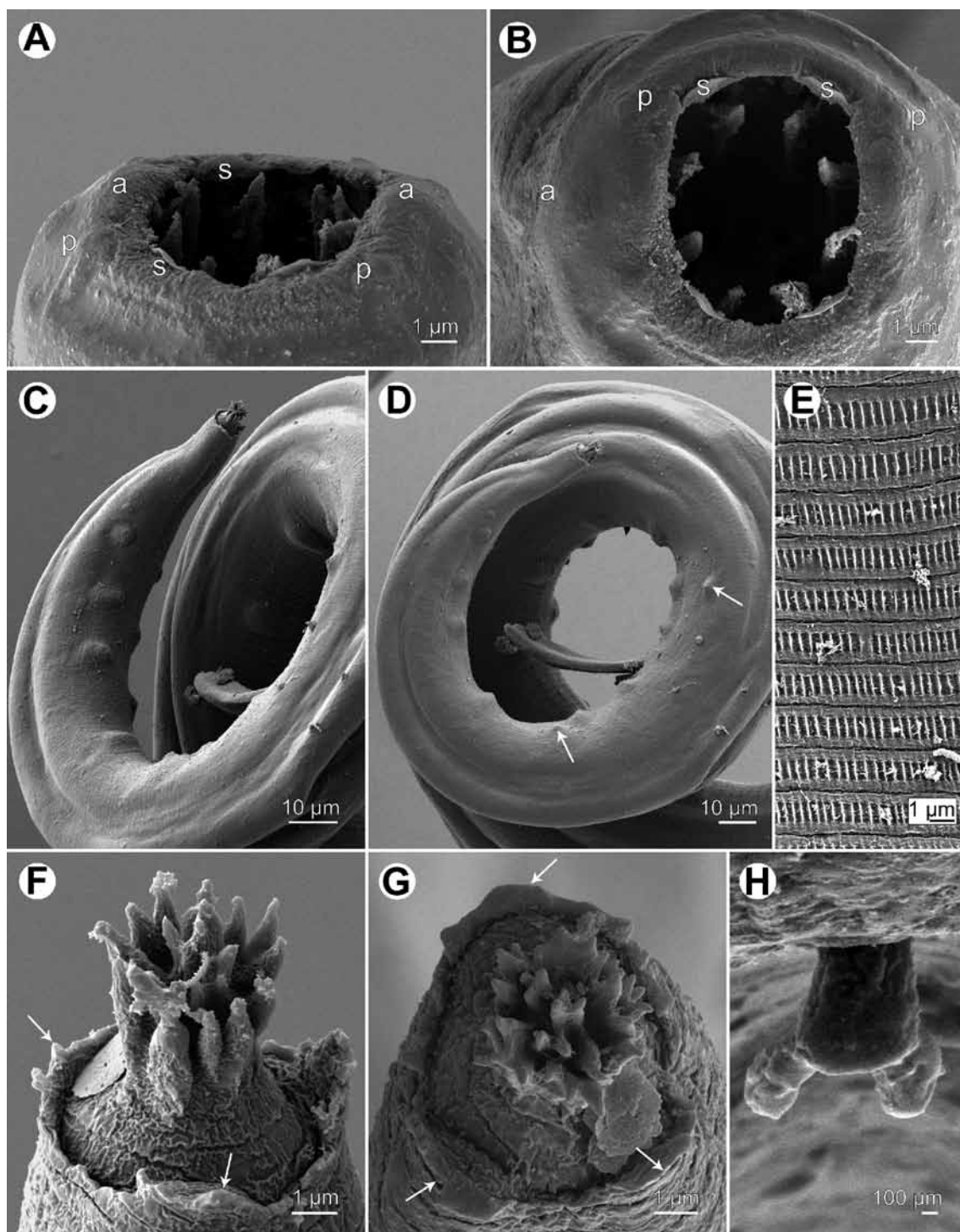


Fig. 2. *Rhabdochona (Globochona) rasborae* sp. n., scanning electron micrographs. **A, B** – cephalic end, lateral and apical views; **C** – tail of male, subventral view; **D** – posterior end of male, lateral view (arrows indicate lateral preanal and postanal papillae); **E** – ventral precloacal cuticular ornamentation of male; **F, G** – tail tip of male, ventral and apical views (arrows indicate thickenings of cuticular ring surrounding central spiny outgrowth); **H** – deirid. *Abbreviations:* a – amphid; p – cephalic papilla; s – sublabium.

Remarks. The genus *Rhabdochona* includes a large number of species parasitizing the intestine of freshwater fishes in all zoogeographical regions (Moravec 2010). At present, 96 species of this genus can be considered to be possibly valid (Moravec et al. 2012a,b). An additional

three species, viz. *R. annai* Kakar, Bilqees et Khan, 2012, *R. bifurcatum* [sic] Kakar et Bilqees, 2012 and *R. pakistana* Kakar, Bilqees et Khan, 2012, have recently been described from fishes in Pakistan (Kakar and Bilqees 2012, Kakar et al. 2012a,b). However, considering their poor

and evidently confused descriptions not corresponding to the requirements of the present nematode taxonomy, all of them should be taken for *species inquirendae*.

Based on the number (8) of anterior prostomal teeth, absence of lateral alae and the presence of caudal projections on the tip of the tail, nematodes of the present material can be assigned to the subgenus *Globochona*, as re-diagnosed by Moravec (1975). Only a few species of *Rhabdochona* (*Globochona*) are known from freshwater fishes in tropical and subtropical Asia and Africa (Moravec 1975, 2010, Boomker and Petter 1993, Wang et al. 1994, Moravec et al. 2010, 2012a, Moravec and Yooyen 2011a). *Rhabdochona* (*Globochona*) *bagarii* Wang et Guo, 1983 proved to be a homonym to *Rhabdochona bagarii* Gupta et Srivastava, 1982 [*species inquirenda*] and, therefore, it was renamed *R. wangi* by Moravec and Scholz (1991).

Of the *Rhabdochona* (*Globochona*) species, only the following six are characterized by the eggs provided with conspicuous lateral swellings (floats): *R. (G.) bariliusi* Soota et Sarkar, 1981 from *Barilius bendelisis* (Hamilton) in India; *R. (G.) barusi* Majumdar et De, 1971 from *Barilius* sp. in India; *R. (G.) gambiana* Gendre, 1922 from an unidentified fish, *Barbus eutaenia* Boulenger and *Raïmas moori* (Boulenger) in Africa (Gambia and the Democratic Republic of Congo); *R. (G.) singhi* Ali, 1956 from *Glossogobius giuris* (Hamilton) in India; *R. (G.) rahimi* Ghazi, Nisa et Bilkees, 2003 from *Barilius pakistanicus* Mirza et Sadiq in Pakistan; and *R. (G.) wangi* Moravec et Scholz, 1991 from *Bagarius bagarius* (Hamilton) and *Zacco platypus* (Temminck et Schlegel) in China (Ali 1956, Majumdar and De 1971, Moravec 1972, Soota and Dey Sarkar 1981, Wang and Guo 1983, Ghazi et al. 2003).

However, in contrast to the all above-mentioned six species, *R. rasborae* sp. n. is noted for the presence of a characteristic, well-developed spinose formation at the tail tip of both the male and female, whereas a similar spinose caudal formation is present on the tail tip of female but absent in the conspecific male in *R. barusi* and *R. wangi*; the female tail tip of *R. gambiana* is covered by many minute cuticular points, but that of the male is rounded and smooth (Moravec 1972); the tail tips of both sexes are simple, without any outgrowths, in *R. bariliusi*, *R. ghazi* and *R. singhi*.

In addition, *R. bariliusi*, *R. barusi* and *R. gambiana* are reported to have their eggs provided with 4 (vs. 2–3) equatorial swellings. The body lengths of gravid females with mature eggs are comparable with that of the new species only in *R. bariliusi* (11.9–13.5 vs. 8.6–23.7 mm) and *R. wangi* (8.3–13.6 mm), whereas the females of the remaining species are distinctly shorter, not exceeding 7.5 mm (mostly less than 6 mm). Moreover, unlike the new species, *R. barusi* has nine (vs. six) pairs of postanal papillae and the distal end of its small spicule is bifurcated, whereas *R. wangi* is reported to possess seven pairs of asymmetrically arranged postanal papillae.

DISCUSSION

Data provided by Kumchoo (2000), Wongsawad et al. (2004) and Boonchot and Wongsawad (2005) have indicated that *Rhabdochona* spp. are common parasites of fishes in Thailand, occurring there in many fish host species belonging to different families. Consequently, it can be assumed that there is a high degree of the species diversity of these nematodes in this zoogeographically very interesting and ecologically varied, but hitherto little explored region. To date, including results of this paper, three nominal species of *Rhabdochona*, *R. (R.) pseudomysti*, *R. (G.) rasborae* and *R. (G.) thaiensis*, have been recorded from Thailand (Moravec and Yooyen 2011a, present study), all representing forms endemic to this country. However, it can be expected that many other congeneric species will be discovered and described in Thailand.

It is worth mentioning that the recently described Thai species of *Rhabdochona* exhibit some unique or rare morphological features within this genus, such as the shape of deirids in *R. pseudomysti* or caudal structures in *R. rasborae* and *R. thaiensis*. A remarkable feature of *R. (G.) rasborae* is the structure of the male ventral pre-cloacal ornamentation (area rugosa). As mentioned by Moravec et al. (2012c), in some *Rhabdochona* (*Rhabdochona*) spp. this male ventral cuticular ornamentation is formed, similarly to some cystidicolids, by several longitudinal ridges or is completely missing. In contrast, in *R. (G.) rasborae* the area rugosa is represented by many cuticular transverse bands bearing numerous short longitudinal ridges (Figs. 1K, 2E). To date, a similar structure of the area rugosa has been reported only for the recently described species belonging to the same subgenus, *Rhabdochona* (*G.*) *kurdistanensis* Moravec, Bilal et Abdullah, 2012 from *Luciobarbus kersin* (Heckel) (Cyprinidae) in northern Iraq (Moravec et al. 2012a) and the African species *R. (G.) paski* Baylis, 1928 (Puylaert 1973, Moravec, unpublished).

The morphology of *R. (G.) rasborae* is also remarkable for the presence of a “spinose formation” at the tail tip of both sexes. Numerous small cuticular outgrowths at the tail tip of females or of both sexes are characteristic mainly of some *Rhabdochona* species of the subgenera *Globochona* and *Globochonoides* Moravec, 1975, but also of one species (*R. salgadoi* Caspeta-Mandujano et Moravec, 2000 in Mexico) belonging to the nominotypical subgenus *Rhabdochona* (see Caspeta-Mandujano and Moravec 2000, Moravec 2010). Because of the small size of these caudal cuticular structures, it is almost impossible to study them in detail by LM. However, recent SEM studies of these caudal formations in a few species of *Rhabdochona* (see Caspeta-Mandujano and Moravec 2000, Moravec and Yooyen 2011a, Moravec et al. 2012a,c, present study) have shown substantial interspecific differences in their structure.

Only two ventral claw-shaped projections are present on the tail tip of *R. (G.) thaiensis*, two lateral denticulate outgrowths, each bearing four horn-like projections, are found on that of *R. (G.) kurdistanensis* (see Moravec and Yooyen 2011a, Moravec et al. 2012a), and the female tail tip of *R. (G.) coronacauda* Belous, 1965 bears two broad lateral outgrowths, each with 4–8 digit-like projections surrounding a smooth central area demarcated by both outgrowths (Moravec et al. 2012d). The female tail tip of *R. (R.) salgadoi* is provided with a small terminal rose-shaped outgrowth bearing numerous digit-like projections. In contrast, the male and female tail tips of *R. (G.) rasbora* have a somewhat more complex structure, where the terminal rose-shaped outgrowth bearing numerous digit-like projections is surrounded by a ring of elevated, thickened cuticle appearing in LM as two small spines in lateral view (Figs. 1N,O, 2F,G). As mentioned by Moravec et al. (2012d), the detailed structure of such a caudal formation on the tail tip may serve as an important taxonomic feature in some *Rhabdochona* spp.

Various aquatic insects (largely ephemeropterans and less often trichopterans and plecopterans) were found to serve as intermediate hosts for some *Rhabdochona* spp. (see Anderson 2000, Moravec 2010), but the life cycles of the representatives of this genus occurring in Thailand remain unknown. Therefore, observations on the transmission of these parasites in natural conditions as well as experimental studies on the life cycles and larval morphogenesis of Thai species of *Rhabdochona* are highly needed.

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