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The first report of new species: *Trichuris landak* n. sp

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PEER REVIEW

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Comments

This paper reported the new species of *Trichuris*–*T. landak*, and compared its appearance structure with other known *Trichuris* sp. using SEM method. It was showed that *T. landak* n.sp share similar rod bands with pathogenic nematodes *T. trichiurus*. This new nematode is the world's first report, the new finding is very important, because *Trichuris* parasitize not only in pigs and *Hystrix*, but also in human and other primates, causing zoonosis–whipworm disease. Further studies, i.e. DNA analysis, vulva surface and infectivity should be launched. This paper should draw infectious disease scholars and the doctor's attention and concern.

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ABSTRACT

Objective: To study nematode parasites morphology of *Hystrix javanica* (*H. javanica*), both through the feces and internal organs. **Methods:** Feces were observed by direct smear method, internal organs were observed after dissecting the host. Specimens for light microscopy examination were fixed with 70% warm alcohol, cleared and mounted in lactophenol for wet mounting. Specimens for SEM examination were postfixed in cacodylate buffer and glutaraldehyde, dehydrated through a graded series of alcohol and freeze dried. The specimens were attached to stubs with double cello–tape, coated with gold and observed with a JSM5310 LV electron microscope. Figures were made with the aid of a drawing tube attached to Olympus compound microscope, other figures were photographs of scanning electron microscope images. Measurements were given in micrometers as the mean followed by the range in parentheses, unless otherwise stated. **Results:** The nematode species found in the intestine of *H. javanica* are *Gireterakis girardi* and a new species, *Trichuris landak*. The new species differs with previously reported species from *Hystrix* because of having stylet and short cervical alae. The pattern of bacillary band is closed to *Trichuris trichiurus*, the species that infect human, but differs because the surface of its vulva is not covered with densely spine. **Conclusions:** The species of nematodes found on *H. javanica* were *Gireterakis girardi* and a new species *Trichuris landak* n.sp. Those two species are newly recorded in Indonesia.

KEYWORDS

Hystrix, Javan porcupine, *Gireterakis girardi*, *Trichuris landak* n. sp., Java, Indonesia

1. Introduction

Javan porcupine, *Hystrix javanica* (F. Cuvier, 1823) (*H. javanica*) usually called landak in Indonesian language, is a small mammal that can be easily recognized due to the spinny surface of its body. Some people in Indonesia believe that the meat and quills of *Hystrix* are usefull for medicinal, aphrodisiac and for ornamental purposes. The number of these animals is getting decrease because they are caught

from nature for illegal trading commodity. To resolve this problem, the Zoological Division of Research Centre on Biology–LIPI, Cibinong have captured the Javan *Hystrix* and built captivity to breed and protect this animal. The biological characteristics of *Hystrix* and breeding technique could be studied through the captivity to meet the need of *Hystrix* for trading commodity. By building the captivity and breeding center, the exploitation of *Hystrix* from natural habitat could be avoided.

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During maintenance of the captive Javan *Hystrix*, the nematode parasites were observed by examining the feces and the internal organs after dissecting dead animals. The information of nematode parasites of Javan *Hystrix* is the basic data that could support the success captivity, and help to find out the role of the parasite to human health, because some nematode species of wild animals are zoonoses[1].

Two species of nematode were found in the intestine of Javan *Hystrix* in captivity, i.e. *Gireterakis girardi* and *Trichuris landak* n. sp. *Gireterakis* is synonymy with *Heterakis*[2], but both are different because *Gireterakis* having straight and short cordon[3]. The species *Gireterakis girardi* (*G. girardi*) has been found in *Hystrix bengalensis* and *Hystrix cristata* (*H. cristata*) in India[2]. *Trichuris* is a kind of parasites in mammals, it is synonymy with *Trichocephalus*[4,5]. The species reported from *Hystrix* in India are *Trichuris infundibulum* and *Trihuris hystricis* (*T. hystricis*) from *H. cristata*[4]. A case of *Trihuris* in human has been reported in a 4 years old child, infected by whipworm (*Trichuris vulpis*) of dog in Maryland[6]. The new record of *Gireterakis* and new species of *Trichuris* were described herein.

2. Materials and methods

Materials used in this study were feces of porcupine from several places in Indonesia and nematode collection from the internal organs after dissecting the dead porcupine. Feces of porcupine from: Central Java (2), Sumatera (1), East Java (1) and West Java (3), were examined by the direct smear methods. Nematode specimens for light microscopy were fixed in 70% warm alcohol, cleared and mounted in glycerine–70% alcohol for examination as wet mounts. Specimens for SEM examination were postfixed in cacodylate buffer and glutaraldehyde, dehydrated through a graded series of alcohol and freeze dried. The specimens were attached to stubs with double cello–tape, coated with gold and observed with a JSM5310 LV electron microscope. Figures 1–14 were made with the aid of a drawing tube attached to Olympus compound microscope, other figures were photographs of scanning electron microscope images. Measurements were given in micrometers as the mean followed by the range in parentheses, unless otherwise stated.

3. Results

The eggs of nematode parasites were found in all feces examined, except one sample from East Java. The eggs were described as *Trichuris* because the specific morphology and those fits with the finding adult worm from the dissecting dead porcupine. Other eggs were believed as eggs of *Gireterakis* as revealed by the findings of adult *Gireterakis*.

The morphology and description of the nematode were presented below.

3.1. Description

3.1.1. *G. girardi* Lane, 1917

Host: *H. javanica*

Site: intestine

Locality: Indonesia

Others host: *Hystrix bengalensis* and *H. cristata*
Distribution: India, Indonesia

3.1.2. General morphology discription

Stout worm, medium in size, cuticle with transversal striation. Mouth with a dorsal lip, and two lateral lips with amphid on each of them (Figures 3 and 15), three papillae present between two cordons, posterior to each lips (Figures 16, 17 arrow). The anterior end of each lips forming straight cordons extending posteriorly at about 150 μ m length (projecting downwards) (Figures 2, 3, 16c and 17c). Lateral alae beginning from behind the lips, extending almost to the length of body, narrow at first, widened at level of nerve ring to esophageal bulb. Esophagus cylindrical, slender, wider at 2/3 of its length, ending in an elongated large bulb are shown in Figure 1.

Figure 1–14. *Gireterakis girardi* (1–8) and *T. landak* n. sp. (9–14).

1. Anterior end of male, ventral view; 2. Head, ventral view; 3. Mouth, en face view; 4. Tail, ventral view; 5. Vagina vera, lateral view; 6. Posterior end of female, lateral view; 7. Spicule; 8. Egg; 9. Anterior end male, ventral view; 10. Female tail, lateral view; 11. Male tail, lateral view; 12. Tip of spicule, lateral view; 13. Egg; 14. Tip of spicule sheath with swelling.

In male *G. girardi* ($n=6$), the body length: 20.284 (19.110–21.640) mm, width at anterior end: 92 (104–115) μ m, maximum width: 772 (710–880) μ m, interval of transversal striation: 7.5 (7.1–7.8) μ m, length of cordon: 148 (135–148) μ m, nerve ring: 175 (158–200) μ m, excretory pore: 747 (620–800) μ m from anterior end. Pharynx: 125 (120–132) μ m, total esophagus length: 1796 (1745–1890) μ m. Tail tapering, short, abruptly narrower at a half length (Figure 4), ventral surface of area in front of the spike clearly striated with ridges (Figures 18 and 19), spike prominent (Figure 18), length: 564 (510–610) μ m. Caudal alae extending from anterior of preloacal sucker to almost the end of tail (Figure 4), united ventrally behind the

Figure 15–27. *G. girardi* (15–20) and *T. landak* n. sp.

15. Mouth, en face view; 16. Lateral lip showing 3 papillae (arrow) and the cordon (c); 17. Papillae (arrow), and the cordon (c); 18. Tip of male tail showing striae on the ventral surface, ventro-lateral view; 19. Magnification of striae no 18; 20. Male tail showing precloacal sucker and caudal papillae; 21. Male tail, showing spicule sheath, lateral view; 22. Anterior part of spicule sheath; 23. Posterior part of spicule sheath, showing dense spine on the surface; 24. Bacillary gland (arrow), anterior part of male; 25. Bacillary gland (arrow), posterior part of male; 26. Pattern of bacillary band (bb); 27. Vulva of female, ventral view.

sucker, wide at level of 1/3 anterior of its length, narrower posteriorly. Caudal papillae 15 pairs: two pairs large, around the preanal sucker, one pair smaller between them, two pairs around the anal, one pair is the largest, fleshy, lateral to anal, one pairs in front of anal, four pairs postanal (Figure 20). Spicule long, forming an angle at 1/3 proximal, narrower to distal part, bent ventrally at the tip, with elongated knob (Figure 7), length: 1620 (1400–1810) μm , preanal sucker with chitinous rim (Figure 20): 138 (130–145) μm in diameter, 853 (780–910) μm from posterior end.

In female ($n=9$), the body length: 23.940 (23.270–24.610) mm, width at anterior end: 135 (130–140) μm , maximum width: 735 (660–810) μm , interval of transversal striation: 7.3 (7.0–7.6) μm , length of cordon: 153 (121–186) μm , nerve ring: 595 (580–610) μm , excretory pore: 750 (720–780) μm from anterior end, pharynx 130 μm , total esophagus length: 1840 (1800–1880) μm . Tail short, tapering, conical end (Figure 6), length was 1720 (1680–1760) μm . Vulva was protruding outward, 6990 (6610–7370) μm from anterior end, vagina running posteriorly (Figure 5) from anterior end. Egg was large; oval, thick shelled (Figure 8), 62 (60–63) by 43 (40–45) μm .

3.1.3. *Trichuris landak* n. sp. (*T. landak*)

Body consists of filiform, longer, smaller oesophageal part and stout, shorter, wider posterior part. Cuticle with transversal striation (Figures 24, 25 and 26s), except for the bacillary band region, mouth with rounded end, stylet arising from the mouth cavity, short cervical alae presents behind the cephalic end, crescent shaped (Figure 9). Bacillary band consists of rectangular protrusion, arranged in longitudinal row, beginning from behind cephalic end to oesophago-intestinal junction; occupy about 1/3 of body width (Figure 26bb). Bacillary gland dense at the anterior part (Figure 24,

arrow) and decreasing to posterior part (Figure 25, arrow), cup shaped, vary in size. Both male and female with bluntly rounded posterior end (Figures 10 and 11). Oesophagus long almost as long as the esophageal part. Nerve ring behind the cervical alae (Figure 9).

Male: Body length was 39.345 (36.050–42.640) mm, esophageal part: 22.385 (21.120–23.650) mm, ratio anterior to posterior part was (1.3–1.4):1, width at posterior end of oesophageal part: 286 (282–290) μm , maximum width of posterior part: 740 (710–770) μm . Cervical alae length: 21 (19–23) width 8.6 (7.8–9.4) μm . Spicule single, slender, attenuated at proximal end forming a knob, slightly narrowed distally, funnel shaped, conical tip (Figures 11 and 12), length 398 (330–465) μm width 44 (38–50) μm . Spicule sheath tubular including the tip but in some specimens the tip is swelling (Figure 14), the surface covered by regular spine, its shorter and less dense at the proximal part (Figure 22), and more dense at the distal part (Figure 23), length of sheath: 398 (330–465) μm , width: 44 (38–50) μm . Cloaca terminal, posterior end spirally coiled (Figure 11).

Female: Body length was 36.390 (35.990–41.912) mm, oesophageal part was 23.095 (22.200–23.790) mm, ratio oesophageal part to posterior part (1.5–1.7): 1, width at posterior end of oesophageal part was 305 (300–310) μm , maximum width of posterior part: 805 (800–810) μm . Cervical alae length: 24 (23–23) μm . Vulva: 18415 (14130–22900) μm from anterior end, cuticle rugose at around the anterior lip (Figure 27), vagina running anteriorly. Egg ellipsoidal with a plug at each poles (Figure 13), measurement of 53 (45–60) by 23 (20–25) μm . Posterior end bluntly rounded, anus subterminal (Figure 10).

3.1.3.1. Remark

T. landak had short lateral alae and stylet, and characters were found in some *Trichuris* species, such as *Trichuris muris* (Shrank, 1788; Hall, 1916), *Trichuris ovis* (Abilgaard, 1795; Smith, 1908) and *Trichuris parvispicularis* (Clapham, 1945; Skrjabin, 1957). The *T. landak* differs from *Trichuris muris* and *Trichuris ovis* because of lateral alae. *Trichuris muris* and *Trichuris ovis* have stylet but no lateral alae. *Trichuris parvispicularis*, parasite of *Thryonomys swinderianus* in Africa also has lateral alae, but differs from *T. landak*, because it does not have stylet, and the body is tightly twisted into intertwining spiral. Pattern of bacillary bands of *T. landak* (Figure 26bb) is closed to *Trichuris trichiurus* (*T. trichiurus*)^[7], the species that could infecting man, but the surface of its vulva is covered with densely spine, *T. landak* is without such structure. It differs from two *Trichuris* species found in *Hystrix*: *Trichuris infundibulus* and *T. hystricis* because it has neither lateral alae nor stylet, and it also differs from *T. hystricis*, because it has no bacillary band^[4], posterior end of spicular sheath with bulb like structure, and have longer spicules (896 *vs.* 398). The new species further differs from *Trichuris infundibulus* in having longer spicules (1940 *vs.* 398). Those differences justify to the conclusion that *T. landak* is a new species.

4. Discussion

Specimens of *G. girardi* reported in the present study do not differ in characteristics and measurements from that previously described, except the anterior somatic papillae. Previous description did not mention papilla, but in the specimens of present study there are three papillae between the cordon, posterior to each lips (Figures 16 and 17 arrow). This characteristic is probably missed from previous observation or it does not exist in the previous specimens described. *G. girardi* has been found in three species of *Hystrix*, and recently there is no report that is from other species of host rather than *Hystrix* spp. The *G. girardi* found in Indonesia is a new record for locality and host species.

Individual characteristic that characterizes *Trichuris* species is indistinct^[4], however, it could be distinguished by looking to the length of spicule, structure of spicular sheath, egg measurements, and length of esophagus. One of the general characteristics of *Trichuris* is spicular sheath, but Artyukh considered that this characteristic was not constant^[8], it changes according to the physiological conditions, therefore, it cannot be used as a standard for distinguishing species. The changes in spicular sheath possibly are in its shaped. Some specimens of *T. landak* have sheath without bulb (Figure 11), and others have tubular sheath with bulb (Figure 14). This fact also occurred in *T. trichiurus* and *Trichuris discolor* (Linstow, 1906). The spicular sheath of *Trichuris infundibulum* has transferred folds proximally, but this structure is not constant^[6]. However, pattern of sheath surface, spinny or not and the relative length of spicules can be used to distinguish the species.

Conflict of interest statement

I declare that I have no conflict of interest.

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General comment

This paper reported the new species of *Trichuris*–*T. landak*, and compared its appearance structure with other known *Trichuris* sp. using SEM method. It was showed that *T. landak* n.sp share similar rod bands with pathogenic nematodes *T. trichiurus*. This new nematode is the world's first report, the new finding is very important, because *Trichuris* parasitize not only in pigs and *Hystrix*, but also in human and other primates, causing zoonosis–whipworm disease. Further studies, *i.e.* DNA analysis, vulva surface and infectivity should be launched. This paper should draw infectious disease scholars and the doctor's attention and concern.

Editorial note

Our journal's main focus is to report the new species, new findings and the latest progress in tropical regions. This first discovery reported *Trichuris landak* n. sp. is of considerable significance to the further research and applications. Today, there are many uncognitive species in tropical regions, APJTB welcomes authors to submit more manuscripts about these kinds of new findings. We will publish the new discoveries and new progress in the first edition in the first place.

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