



Revision of tapeworms (Cestoda: Bothriocephalidea) from lizardfish (*Saurida*: Synodontidae) from the Indo-Pacific region

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Abstract

Bothriocephalidean cestodes reported from lizardfish (*Saurida* Valenciennes in Cuvier & Valenciennes) were revised on the basis of newly collected material from type-hosts and available type- and voucher specimens. Instead of nine taxa of four genera listed in the literature, only *Oncodiscus sauridae* Yamaguti, 1934 and *Penetrocephalus ganapattii* (Rao, 1954) are considered valid and thus both genera become monotypic. Both taxa are redescribed, including the first scanning electron micrographs and data on intraspecific variability. Both species are unique among bothriocephalid cestodes in the lateral position of the vagina in relation to the cirrus-sac. *Oncodiscus sauridae* found in *Saurida longimanus* Norman, *S. nebulosa* Valenciennes (new host record), *S. tumbil* (Bloch) and *S. undosquamis* (Richardson) differs from *P. ganapattii* found in *S. micropectoralis* Shindo & Yamanda and *S. tumbil* in the following characters: (i) scolex shape (fan-shaped, with crenulated bothrial margins in *O. sauridae*, replaced by a *scolex deformatus* in *P. ganapattii*); (ii) site of scolex attachment (in the intestinal lumen in the former taxon versus penetrating through the intestinal wall, and encysted on pyloric caeca in *P. ganapattii*); (iii) shape of segments (usually only slightly wider than long in *O. sauridae* versus very short, much wider than long in *P. ganapattii*); and (iv) number of testes (50–100 per segment in *O. sauridae* whereas fewer than 60 in *P. ganapattii*). *Bothriocephalus sauridae* Ariola, 1900 is considered a *species inquirendum*, although conspecificity with *O. sauridae* is very probable. The present study demonstrates the necessity of using standard procedures, especially adequate fixation methods of freshly collected cestode material (hot 4% formaldehyde solution is strongly recommended for morphological studies), because most previous descriptions were based on poor-quality material, including partly macerated worms missing hooklets on the apical disc of the scolex in *O. sauridae*.

Key words: Bothriocephalidae, Eucestoda, Indo-Pacific, Pseudophyllidea, lizardfish, *Oncodiscus sauridae*, *Penetrocephalus ganapattii*, *Saurida*, Synodontidae, tapeworm

Introduction

Fish parasitological studies in the Indo-Pacific region have indicated that several species have a wide range of distribution throughout the region, causing taxonomic confusion due to duplicate species descriptions (e.g. Palm & Overstreet 2000). Palm (2000, 2004) summarized the trypanorhynch cestode fauna from the southern Indonesian coast, demonstrating significant overlap of species composition in Indian and southern Indonesian waters. Lönnberg (1893) described the first tetraphyllidean cestode from Java, and Yamaguti (1954) studied

tetraphyllidean and trypanorhynchan cestodes from the southern Sulawesi coast. However, other information on cestodes parasitic in fish especially from the Indonesian archipelago, a centre of marine biodiversity, is scarce.

Lizardfish of the genus *Saurida* Valenciennes in Cuvier & Valenciennes (Aulopiformes: Synodontidae) are common on the Indo-West Pacific continental shelf, with 21 species recognised as valid (Froese & Pauly 2008). As many as nine nominal cestode species of four genera have been described from four species of lizardfish (Kuchta and Scholz 2007; Kuchta *et al.* 2008b), namely *Bothriocephalus sauridae* Ariola, 1900; *B. ganapattii* Rao, 1954 [= *Penetrocephalus ganapatii* (Rao, 1954) Rao, 1960]; *Bothriocephalus indicus* Ganapati & Rao, 1955; *Bothriocephalus penetratus* Subhadrappa, 1955; *Oncodiscus sauridae* Yamaguti, 1934; *O. fimbriatus* Subhadrappa, 1955; *O. waltirensis* Shinde, 1975; *O. maharashtrae* Jadhav & Shinde, 1981; and *Tetrapapillocephalus magnus* Protasova & Mordvinova, 1986.

The genus *Oncodiscus* Yamaguti, 1934 was revised by Khalil & Abu-Hakima (1985) on the basis of material collected from *Saurida undosquamis* (Richardson) from Kuwait Bay and Australian waters. The authors synonymised two species, *O. fimbriatus* Subhadrappa, 1955 and *O. waltirensis*, both found in *Saurida tumbil* (Bloch) from the Bay of Bengal off India, with *O. sauridae* described from *Saurida argyrophanes* (Richardson) (syn. of *S. tumbil*). These synonymies were accepted by Kuchta & Scholz (2007) and Kuchta *et al.* (2008b), who presented a preliminary list of valid taxa of bothriocephalideans, including new synonymies of species parasitic in lizardfish. However, they did not provide detailed data on their morphology and morphometry.

The purpose of the present study is a revision of bothriocephalidean tapeworms from lizardfish based on newly obtained material from Indonesia and New Caledonia. The morphology and morphometry of the valid species are described, comparing the new material with type and voucher specimens from different museum collections. The re-descriptions of *Oncodiscus sauridae* and *Penetrocephalus ganapattii* including details of the surface ultrastructure are presented in this paper.

Material and methods

The present study was based on morphological and biometrical evaluation of freshly collected specimens as well as on tapeworms deposited in museum collections. New material was isolated from 153 freshly killed lizardfish of 4 species [*S. longimanus* Norman – 52, *S. micropectoralis* Shindo & Yamada – 11, *S. tumbil* – 46, *S. undosquamis* – 44] obtained from local fisherman in Pelabuhan Ratu, Java, Indonesia and examined by the present authors (R.K., A.T.Y., T.W. and H.W.P.) from 2001 to 2008. Other cestodes were collected from 24 lizardfish of 3 species [*Saurida gracilis* (Quoy & Gaimard) – 1, *S. nebulosa* Valenciennes – 1, *S. undosquamis* – 22] in New Caledonia by J.-L. Justine from 2003 to 2006.

Tapeworms collected by the present authors were obtained from freshly killed lizardfish. They were washed in saline and specimens for morphological studies, including scanning electron microscopy (SEM) observations and histology, were fixed with hot 4% formaldehyde solution. Remaining tapeworms or their pieces were fixed with 96% ethanol for molecular study (see Brabec *et al.* 2006). Whole mounts were stained with Schuberg's hydrochloric carmine and mounted in Canada balsam. Cross sections of the strobila (thickness 15 µm) were stained with hematoxylin-eosin, using standard histological methodology (Scholz & Hanzelová 1998). Several scoleces and segments were prepared for SEM following the procedure outlined by Scholz *et al.* (1998).

The following material was studied:

Oncodiscus sauridae

- (i) type material (9 specimens) ex *Saurida argyrophanes* (now *S. tumbil*) from the Inland Sea and East China Sea, Japan (Meguro Parasitological Museum, Tokyo, Japan – acronym MPM): holotype [MPM 23176 (SY 3042)], paratypes [MPM 23176 (SY 3043), MPM 23177 (SY 3165-67)];
- (ii) three specimens ex *Chirocentrus dorab* (Forsskål) from Toyama Bay, Toyama Prefecture, Honshū, Japan, Japan Sea (collected by S. Yamaguti) [MPM 23178 (SY5616-18)];
- (iii) six specimens ex *S. tumbil* from Pelabuhan Ratu, Java, Indonesia (helminthological collection of the Institute of Parasitology, BC AS CR, České Budějovice – IPCAS C-456/1; Natural History Museum, London, UK – BMNH 2008.9.5.3; Invertebrate Collection, Muséum d'Histoire Naturelle, Genève, Switzerland – MHNG INVE 62588);
- (iii) six specimens ex *S. longimanus* from Pelabuhan Ratu, Java, Indonesia (IPCAS C-456/2; BMNH 2008.9.5.6 and 2008.11.7.1; Zoologisches Museum Berlin – ZMB 7447 and 7448);
- (iv) one specimen ex *S. undosquamis* from Pelabuhan Ratu, Java, Indonesia and two specimens from off Nouméa, New Caledonia (IPCAS C-456/3; Muséum National d'Histoire Naturelle, Paris, France – MNHN JNC502, 2588);
- (v) one specimens from *S. nebulosa* from off Nouméa, New Caledonia (MNHN JNC2474).

Tetrapallocephalus magnus

- (i) The holotype ex *S. undosquamis* from the Indian Ocean (Parasitological Institute, Russian Academy of Sciences, Moscow, Russia – GELAN): holotype (GELAN 696);
- (ii) plerocercoid on the slide with the holotype of *Partitotestis berycis* Protasova & Parukhin, 1986 (GELAN 986).

Penetrocephalus ganapattii

- (i) two specimens ex *S. tumbil* from Pelabuhan Ratu, Java, Indonesia (IPCAS C-462/1; MHNG INVE 62589; ZMB 7449).
- (ii) three specimens ex *S. micropectoralis* from Pelabuhan Ratu, Java, Indonesia (IPCAS C-462/2; BMNH 2008.9.5.7–8; MHNG INVE 62590; ZMB 7450).

Illustrations were made using a drawing attachment on the Olympus BX51 microscope with Nomarski interference contrast. Types or vouchers of the taxa described by Indian authors were not available and they most probably do not exist or have never been deposited (B. Jadhav – personal comm.).

Results

Based of the evaluation of types, vouchers and newly collected specimens, two taxa belonging to two monotypic genera, *Oncodiscus sauridae* and *Penetrocephalus ganapattii*, are considered valid. They represent nine nominal species of four bothriocephalidean genera that were formerly described from lizardfish. Both species are redescribed herein, with all measurements are given in micrometers (µm); those of the type material are in parentheses.

***Oncodiscus sauridae* Yamaguti, 1934**

Figs. 1–8, 14–18, 21, 24

Syns. *O. fimbriatus* Subhadrappa, 1955; *O. waltirensis* Shinde, 1975; *O. maharashtrae* Jadhav & Shinde, 1981; *Bothriocephalus indicus* Ganapati & Rao, 1955; *Tetrapapillocephalus magnus* Protasova & Mordvinova, 1986

Type host: *Saurida argyrophanes* (now *S. tumbil*).

Other hosts: *S. longimanus*, *S. nebulosa* (new host), *S. undosquamis*.

Site of infection: Scolex in the pyloric caeca or embedded in the intestinal wall, strobila in the intestinal lumen.

Type locality: Inland Sea, Tarumi-ku, Kobe, Hyōgo Prefecture, Kyūshū, Japan, Pacific Ocean.

Distribution: Indian Ocean (India, Indonesia, Kuwait, Mozambique); Pacific Ocean (Australia, Japan, New Caledonia).

Prevalence: In Indonesia, *S. longimanus* – 12%; *S. tumbil* – 9%; *S. undosquamis* – 5%. In New Caledonia, 9% of *S. undosquamis* were infected (J.-L. Justine – unpublished data).

References: Subhadrappa (1955); Devi (1975); Khalil & Abu-Hakima (1985); Protasova & Mordvinova (1986).

Re-description: Bothriocephalidea, Bothriocephalidae. Medium-sized worms, up to 130 (90) mm long. Strobila with craspedote segments with prominent posterolateral wing-like projections, overlapping following segment (Figs. 5, 16, 17). External segmentation present, but may be incomplete (missing medially). Two pairs of longitudinal osmoregulatory canals present. Longitudinal musculature well developed, formed by numerous bundles of muscle fibres (Fig. 7). Surface covered with small filiform microtriches similar in shape and size.

Scolex oval, irregularly-shaped or fan-shaped (Figs. 1, 2, 4, 14, 15), usually compressed laterally, 1,120–3,140 (1,710–4,000) long by 860–2,380 (1,360–2,190) wide. Bothria well developed, with conspicuously crenulated margins turned laterally (Figs. 1, 2, 4, 14, 15). Apical disc well developed, 310–890 (780–1,250) wide, 140–400 (160–370) high, consisting of two lateral lobes, armed with up to four irregular rows of tiny hooklets (Figs. 2, 3, 15, 18, 24); hooklets easy to detach, especially in dead specimens (Figs. 1, 4), 21–26 (12–24) long, with largest hooklets in second and third rows, and diminishing in size from top of lateral lobes to junction of lobes, where rows may be interrupted. Neck absent, first segments starting immediately posterior to scolex (Figs. 1, 15).

Immature segments much wider than long, 25–110 (45–165) long by 400–1,510 (230–850) wide. Mature segments wider than long, 50–810 (80–380) long by 310–1,715 (755–1,070) wide (Fig. 5). Gravid segments wider than long to rectangular or, exceptionally, slightly longer than wide, 190–2000 (180–700) long by 615–1865 (985–1520) wide (Figs. 16, 17).

Testes medullary, oval, 50–100 (60–100) in number per segment, 22–66 (30–61) long by 11–38 (25–50) wide, in two lateral fields, usually separated medially, continuous between segments (Fig. 5). Cirrus-sac submedian, irregularly alternating, thick-walled, round or slightly oval, 70–215 (85–135) in diameter, just anterodorsal to ovary (Figs. 5, 6, 8). Cirrus armed with filiform microtriches. Cirrus opening to small genital atrium lateral to median line of segment (Fig. 7). Genital pore dorsal, median, slightly postequatorial (Fig. 5).

Ovary transverse elongate, bilobed, lobulate, median, near posterior margin of segment, 200–630 (225–460) wide (Figs. 5, 6). Vagina tubular, dorsal to ovarian bridge, arched lateral or anterolateral to cirrus-sac (Figs. 5, 6). Vitelline follicles numerous, cortical, circumsegmental (Fig. 7), oval, 13–40 (7–28) in diameter, in two lateral fields, often separated medially, reaching to posterolateral projections (Figs. 5, 7). Uterine duct well developed, sinuous, ending in muscular, thick-walled uterine sac situated medially (Figs. 5, 6); sac markedly enlarged in last gravid segments. Uterine pore medioventral, at distance from anterior margin of segment or almost equatorial. In some segments, pore extremely large due to collapse of wall of uterus (Fig.

16). Eggs oval, operculate, unembryonated, 55–64 (60–63) long by 32–45 (39–42) wide, with operculum 11–20 in diameter (Fig. 21).

Remarks: *Oncodiscus sauridae* was described by Yamaguti (1934) from *Saurida argyrophanes* (now *S. tumbil*) and redescribed by Khalil & Abu-Hakima (1985), who collected new material and added morphological data to the original description. The total length of 9 mm was given by Yamaguti (1934) for the holotype, but this specimen measures in fact 90 mm (present study).

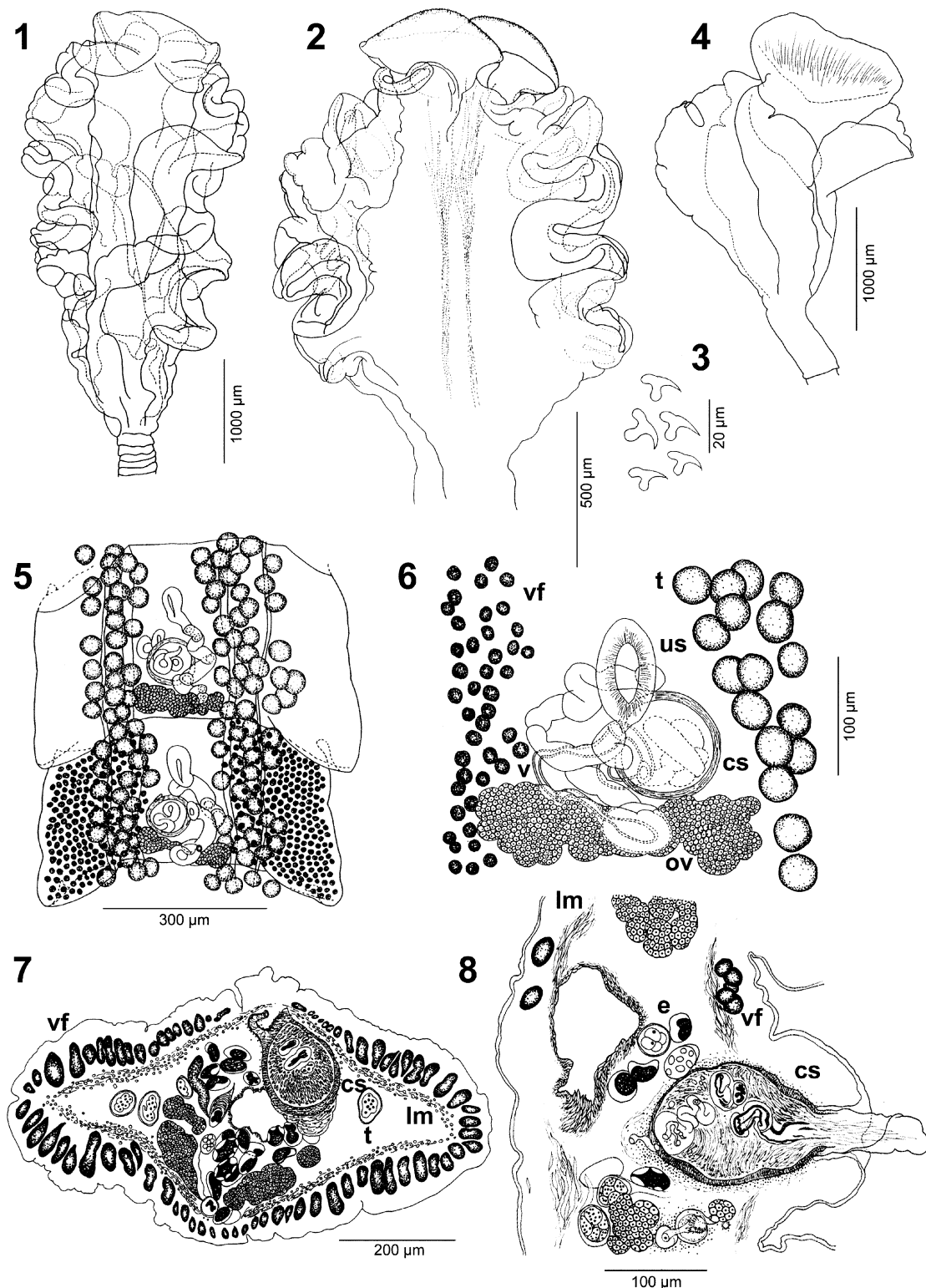
Besides *O. sauridae*, another three species of *Oncodiscus*, namely *O. fimbriatus*, *O. waltirensis*, and *O. maharashtrae*, were erected. They were differentiated mainly by the shape of the scolex, the presence or absence of a median longitudinal furrow, the presence or absence hooklets on the apical disc, the number of testes and the presence or absence of an operculum in the eggs (Subhadrappa 1955; Shinde 1975; Jadhav & Shinde 1981). However, these characters are unsuitable for species differentiation in this group of cestodes. The shape of the scolex and the presence of furrows and hooklets on the apical disc are not stable characters because they depend to a great extent on the state in which worms were fixed and on fixation procedure (Kuchta & Scholz 2007). In dead tapeworms or in worms that have been relaxed for too long in water, the hooklets detach (one of Yamaguti's paratypes of *O. sauridae* lacks hooklets on the apical disc due to tissue destruction – Fig. 1), the scolex shape changes and tegumental furrows may appear as a consequence of deformation of the scolex, especially when fixation by pressure between two glass slides is used (Khalil & Abu-Hakima 1985; present study).

The number of testes is variable in *O. sauridae* and may considerably differ even between segments of the same specimen. The maximum number of testes in *O. sauridae* was found to reach up to 100, which corresponds to the data from the original description (Yamaguti 1934). Jadhav & Shinde (1981) reported as many as 350 testes in one segment of *O. maharashtrae*, but it is apparent that the authors confused vitelline follicles with testes. Vitelline follicles enlarge in the posterior part of the strobila and may reach a similar size compared to that of the testes in gravid segments.

Another feature which was used for species differentiation, i.e. the presence or absence of an operculum in the eggs, can easily be overlooked unless scanning electron microscopy is used (Bray *et al.* 1994; Kuchta *et al.* 2008a; present study). Yamaguti (1934) described a median longitudinal furrow on the scolex of *O. sauridae*. On the basis of the absence of that structure, Subhadrappa (1955) proposed *O. fimbriatus*. However, this structure does not represent a species-distinguishing character. Such a furrow is missing in one of the paratypes of *O. sauridae* and it is much less developed in other specimens of the type series. This indicates that its presence depends on the level of worm contraction or fixation procedure (S. Yamaguti strongly flattened the worms, which may have caused unnatural deformations of the scolex and strobila).

Khalil & Abu-Hakima (1985) synonymized *O. fimbriatus* and *O. waltirensis* with *O. sauridae*, but they considered *O. maharashtrae* to be a *species inquirendum*. This species was described from *Pastinachus (Trygon) sephen* (Forsskål) (Dasyatidae) from off Bombay, India. Unlike Khalil & Abu-Hakima (1985), the present authors (Kuchta & Scholz 2007; Kuchta *et al.* 2008b; this study) consider *O. maharashtrae* to be a synonym of *O. sauridae* which occurred in an unusual (postcyclic or accidental) host.

All three species from India were described on the basis of only a few, apparently macerated and deformed specimens. In addition, the descriptions of the taxa from the Indian Ocean were superficial and no histological sections were made (Ariola 1900; Subhadrappa 1955; Shinde 1975; Jadhav & Shinde 1981; Protasova & Mordvinova 1984). *Bothriocephalus indicus* was described from *Saurida tumbil*. It is obvious from its morphology that the authors erroneously placed these specimens in the genus *Bothriocephalus* Rudolphi, 1808, because species of this genus are devoid of a fan-shaped scolex with crenulated bothrial margins (Ganapati & Rao 1955). In fact, *B. indicus* is identical in its morphology with *O. sauridae* (the absence of hooklets on an apical disc is apparently due to poor state of specimens studied), parasitizes the same species of fish hosts and occurs in the same region. Therefore, the synonymy of *B. indicus* with *O. sauridae*, first proposed by Kuchta & Scholz (2007), is confirmed in the present study.



FIGURES 1–8. *Oncodiscus sauridae* Yamaguti, 1934. **1**, Scolex of paratype (MPM SY 3043); lateral view. **2**, **3**, Voucher (MPM SY 5616) ex *Chirocentrus dorab*; **2**, scolex; **3**, detail of hooklets. **4**, Scolex of the holotype of *Tetrapapillocephalus magnus* (GELAN 696). **5**, Schematic drawing of mature segments ex *Saurida longimanus* (IPCAS C-456/2); vitelline follicles illustrated only in the posterior segment; ventral view. **6**, Detail of mature segment ex *S. tumbil* (IPCAS C-456/1); vitelline follicles illustrated only on the left side and testes only on right side, ventral view. **7**, **8**, Histological sections of gravid segments ex *S. tumbil* (IPCAS C-456/1) at level of cirrus-sac; **7**, cross-section; **8**, sagittal section. Abbreviations: cs – cirrus-sac; e – eggs; lm – inner longitudinal muscles; ov – ovary; t – testes; us – uterine sac; v – vagina; vf – vitelline follicles.

Bothriocephalus sauridae was very briefly described on basis of specimens without a scolex that were collected from *Saurida nebulosa* off the eastern coast of Africa (Zanzibar) (Ariola 1900). The species was considered to be a *species inquirendum* (Protasova 1977; Kuchta & Scholz 2007; present paper), but its conspecificity with *O. sauridae* is very probable. In the same region, *O. sauridae* (reported to as *Tetrapapillocephalus magnus*) was found in *S. undosquamis* by Protasova & Mordvinova (1986).

Morphological and morphometrical examination of the specimens of *Tetrapapillocephalus magnus* (holotype and voucher), which are both in poor condition, shows a high similarity with *O. sauridae*. The taxa differ only in the absence of tiny hooklets on the apical disc in the former. However, hooklets can be easily lost during handling or when dead tapeworms are collected (Khalil & Abu-Hakima 1985; present study).

The scolex of *Tetrapapillocephalus magnus* (= *O. sauridae*) illustrated in the original description (Protasova & Mordvinova 1986) belongs to another specimen, not representing the holotype as indicated (Fig. 4). This immature specimen was found in *Beryx splendens* Lowe; it is mounted onto the same slide with the holotype of *Partiotestis berycis* Protasova & Parukhin, 1986 (GELAN 986). Numerous immature tapeworms conspecific with *T. magnus* were found in this host, but only one specimen was mounted (Protasova – pers. com.). Besides this report, no further data exist on the occurrence of *O. sauridae* in berycid fish (Kuchta & Scholz 2007; Kuchta *et al.* 2008b) and it is possible that *B. splendens* represents an accidental or paratenic host only.

It is interesting to note that we examined some unusual material from clupeid fish (*Chirocentrus dorab*) from the Japan Sea, in the collection of Yamaguti (MPM), collected 19.vi. 1928. These specimens are conspecific with *O. sauridae* (also identified as *Oncodiscus* by Yamaguti), but this finding was never published. In addition, no other occurrence of any bothriocephalid tapeworm has been reported from this host. This finding may represent misidentification of the host, or an accidental or paratenic host.

***Penetrocephalus ganapattii* (Rao, 1954) Rao, 1960**

Figs. 9–13, 19, 20, 22, 23

Syns. *Bothriocephalus ganapattii* Rao, 1954; *Bothriocephalus penetratus* Subhpradha, 1955.

Type host: *Saurida tumbil*.

Other hosts: *S. micropectoralis*, *S. undosquamis*.

Site of infection: The scolex and neck (*scolex deformatus*) is coiled within a cyst on the wall of pyloric caeca, whereas the strobila lies freely in the intestinal lumen.

Type locality: Coast of Waltair, Bay of Bengal, Andhra Pradesh, India, Indian Ocean.

Distribution: Indian Ocean (India, Indonesia).

Prevalence: In Indonesia, *S. micropectoralis* – 27%; *S. tumbil* – 5%.

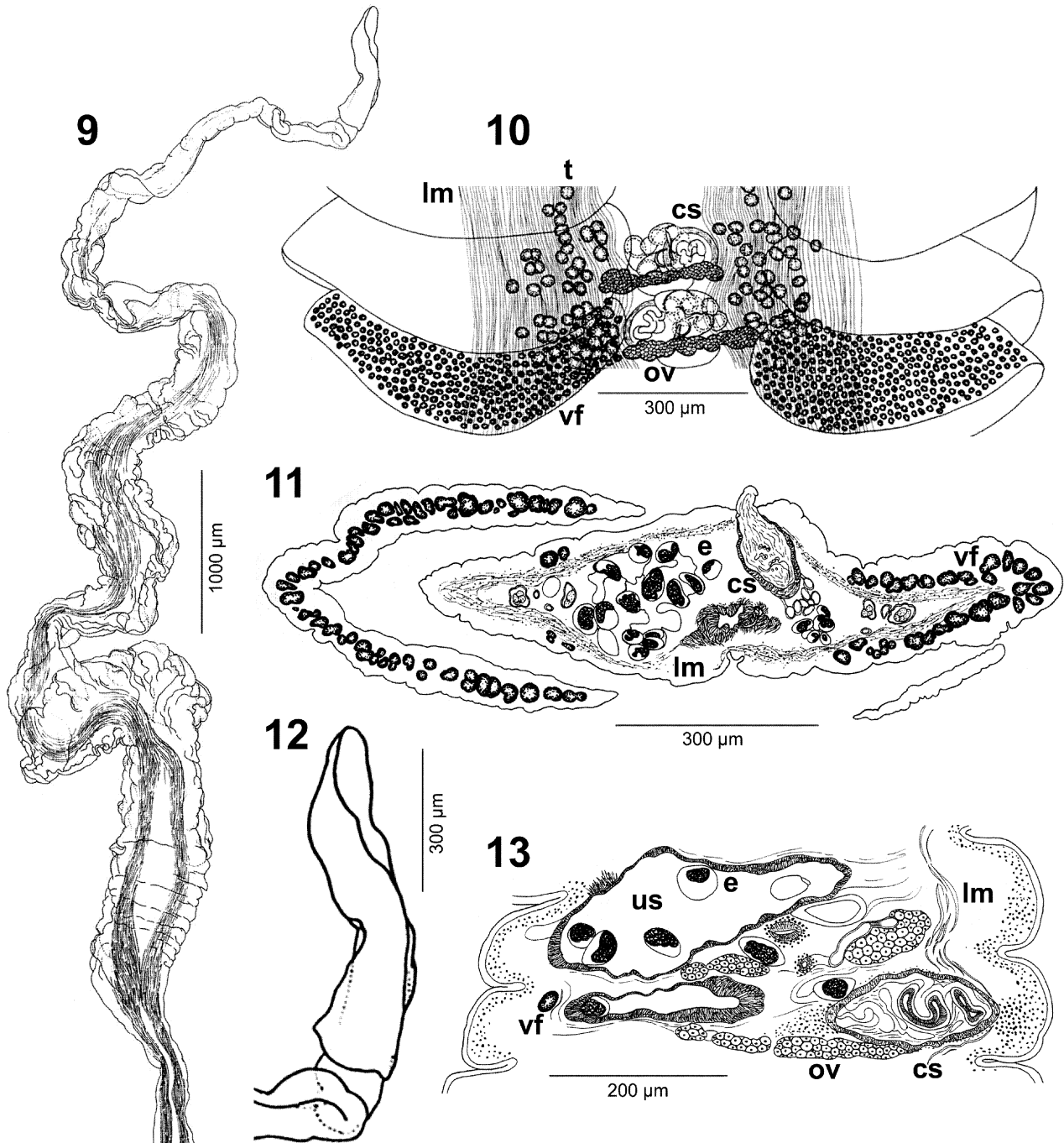
References: Subhpradha (1955); Rao (1960); Devi (1975); Radhakrishnan *et al.* (1983).

Re-description: Bothriocephalidea, Bothriocephalidae. Large worms, up to 125 mm long. Strobila with markedly craspedote segments with prominent lateral wing-like projections (Figs. 10, 19, 23). External segmentation present, may be incomplete (missing medially). Two pairs of longitudinal osmoregulatory canals present. Longitudinal musculature well developed, forming numerous bundles of muscle fibres (Figs. 10, 11). Surface covered with small filiform microtriches similar in shape and size.

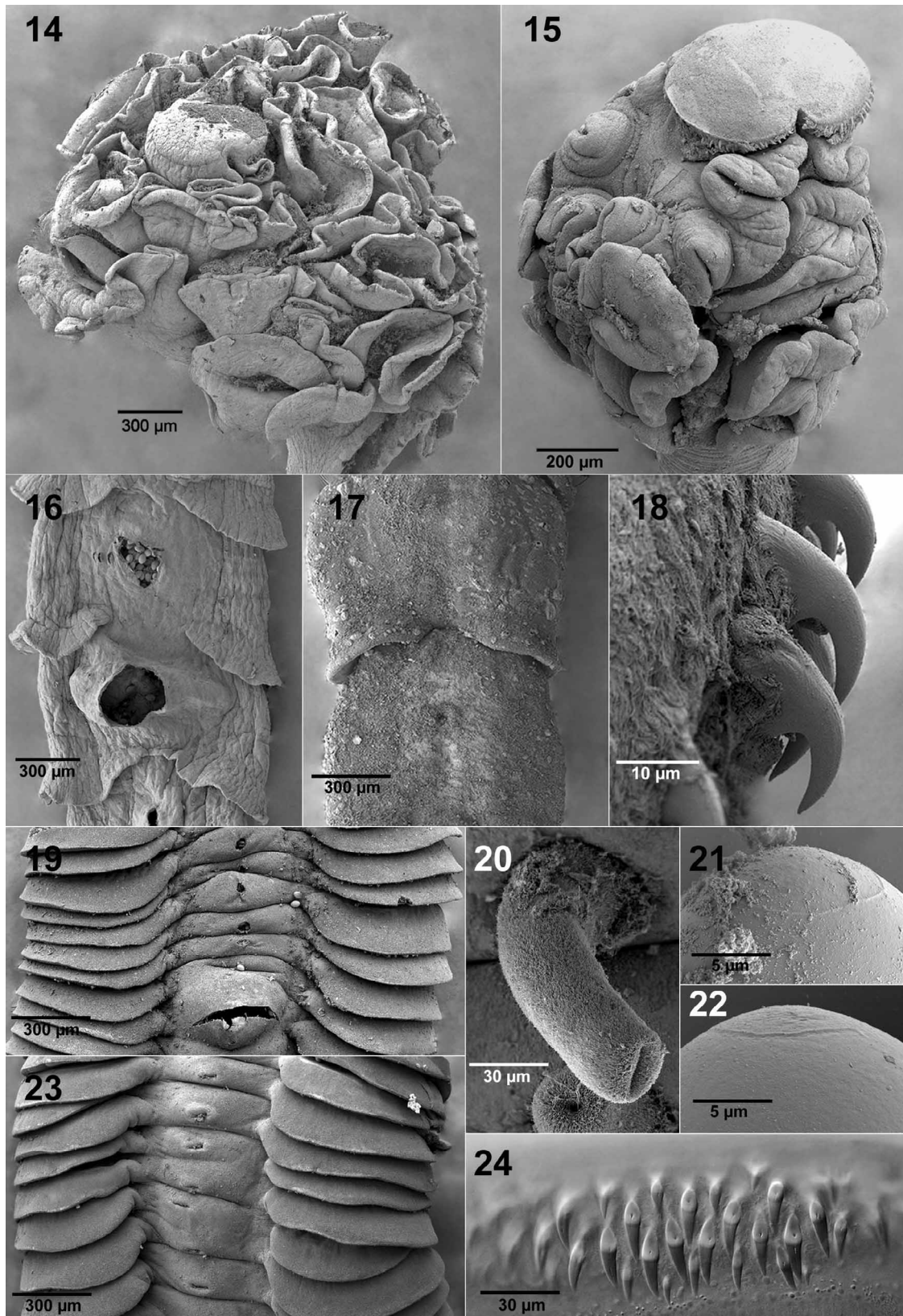
Scolex unarmed, 700–750 long by 150–200 wide, changed to *scolex deformatus* (Fig. 9, 12). Apical disc absent. Anterior part of body including neck often degenerated, becoming filiform (Fig. 9), up to 60 mm long, penetrating through intestinal wall, and encysted on pyloric caeca.

Segments short, much wider than long (up to 14 times in mature segments). Immature segments 50–620 long by 100–2,540 wide. Mature segments 40–220 long by 1,080–2,900 wide (Fig. 10). Gravid segments 70–620 long by 890–3,140 wide (Figs. 19, 23).

Testes medullary, oval, 40–60 in number per segment, 10–60 long by 10–40 wide, in two lateral fields separated medially, continuous between segments (Fig. 10). Cirrus-sac submedian (Fig. 11), irregularly alternating, round or slightly oval, 65–195 in diameter, just anterior to ovary (Fig. 10, 13). Cirrus unarmed, covered with long filiform microtriches (Fig. 20), opening to small genital atrium. Genital pore dorsal, slightly submedian (Figs. 11, 23).



FIGURES 9–13. *Penetrocephalus ganapattii* (Rao, 1954) ex *Saurida micropectoralis* (IPCAS C-462/2). **9, 12**, Schematic drawing of *scolex deformatus*. **10**, Schematic drawing of mature segment; vitelline follicles illustrated only in the posterior segment; ventral view. **11, 13**, Histological sections of gravid segments at the level of cirrus-sac; **11**, cross-section; **13**, sagittal section. Abbreviations: cs – cirrus-sac; e – eggs; lm – inner longitudinal muscles; ov – ovary; t – testes; us – uterine sac; vf – vitelline follicles.



FIGURES 14–23. Photomicrographs. **14–18, 21, 24,** *Oncodiscus sauridae* Yamaguti, 1934. **14,** Scolex; ex *Saurida longimanus*. **15,** Scolex; ex *S. tumbil*. **16, 17,** Gravid strobila; **16,** Ventral view; **17,** Dorsal view. **18, 24,** Detail of hooklets; **18,** SEM photomicrograph; **24,** Photomicrograph. **21,** Operculum of the egg. **19, 20, 22, 23,** *Penetrocephalus ganapattii* (Rao, 1954). **19, 23,** Gravid strobila; **19,** Ventral view; **23,** Dorsal view. **20,** Detailed view of the envaginated cirrus. **22,** Operculum of the egg.

Ovary transverse elongate, lobulate, median, 240–720 wide (Fig. 10). Vagina tubular, lateral or anterolateral to cirrus-sac. Vitelline follicles cortical, oval, 10–40 in diameter, numerous, in two lateral fields separated medially, reaching to posterolateral projections (Fig. 10, 11). Uterine duct sinuous, leading to thick-walled, median uterus (Figs. 10, 13), dilated ventrally in gravid segments. Uterine pore ventral, median, at distance from anterior margin of segment (Fig. 19). Eggs oval, operculate, unembryonated, 50–74 long by 33–42 wide; operculum 9–18 in diameter (Fig. 22).

Remarks: The original description of *Bothriocephalus ganapattii* was very brief (Rao, 1954), which led Protasova (1977) and Bray *et al.* (1994) to consider *B. ganapattii* a *nomen nudum*. However, the species is clearly recognizable on the basis of its original description because it possesses a *scolex deformatus* that penetrates the intestine and forms a cyst in the mesentery or in the liver (Rao 1954). Subhadrappa (1955), probably not aware of Rao's (1954) publication, described an apparently conspecific cestode, *Bothriocephalus penetratus*. Both taxa parasitize the same fish host in the same region and are morphologically indistinguishable. Therefore, they are considered synonymous (Rao 1960; Kuchta & Scholz 2007; Kuchta *et al.* 2008b).

Rao (1960) proposed a new genus, *Penetrocephalus*, to accommodate *B. ganapattii* (the type- and only species was misspelled as *P. ganapatii*). The genus was invalidated by Protasova (1977) and Bray *et al.* (1994), but resurrected by Kuchta *et al.* (2008b); the validity of the genus is also supported by molecular data (Kuchta 2007, fig. 10).

Discussion

The present study contributes to our knowledge of the fish parasite biodiversity from Indonesian coastal waters, a centre of marine biodiversity within the Indo-Pacific region. It confirms the previous conclusions of Khalil & Abu-Hakima (1985) and Kuchta & Scholz (2007) regarding synonymy of bothriocephalid cestodes described from lizardfish. Based on the present data, only two out of nine nominal species are considered valid (*Bothriocephalus sauridae* is kept as a *species inquirendum*). The invalidity of the monotypic genus *Tetrapillocephalus*, as first proposed by Kuchta & Scholz (2007), is confirmed.

Evaluation of freshly collected specimens from Indonesia and New Caledonia has shown high intraspecific and even individual variability in many characters, some of them having been used by previous authors for erection of new taxa (shape of the scolex and apical disc, presence/absence of hooklets on the apical disc and longitudinal furrows on the scolex, measurements of segments, etc.). The present study also demonstrated that poor-quality material, especially macerated cestodes or worms fixed under pressure, should not be used for taxonomic studies in bothriocephalidean cestodes (Kuchta & Scholz 2007).

Oncodiscus sauridae and *Penetrocephalus ganapattii* differ from other bothriocephalid cestodes in the position of the vaginal pore, which is anterolateral to the male pore (opening of the cirrus-sac), whereas in other species of the family Bothriocephalidae it is posterior to the male pore (Kuchta *et al.* 2008b). *O. sauridae* and *P. ganapattii* are also characterized by the shape of segments which possess posterolateral projections, markedly developed mainly in the latter taxon. Similar structures are known only in some echinophallids (Kuchta *et al.* 2008a, b) and several species provisionally placed in *Bothriocephalus*, namely *B. antarcticus* Wojciechowska, Pisano & Zdzitowiecki, 1995; *B. bengalensis* Devil, 1975; *B. branchiostegi* Yamaguti, 1952; and *B. gadellus* Blend & Dronen, 2003 (Protasova 1977; unpublished data).

The species from lizardfish share some morphological characters, especially of the strobila, but they can easily be distinguished from one another. The most important characters are: (i) the scolex shape (fan-shaped, with crenulated bothrial margins (Ganapati & Rao 1955). in *O. sauridae*, whereas replaced by a *scolex deformatus* in *P. ganapattii*); (ii) site of scolex attachment (in the intestinal lumen in the former versus penetrating through the intestinal wall and encysted on pyloric caeca in *P. ganapattii*); (iii) shape of segments (only slightly wider than long in *O. sauridae* versus very short, much wider than long in *P. ganapattii*); and (iv) the

number of testes (50–100 per segment in *O. sauridae* whereas fewer than 60 in *P. ganapattii*). A comparative analysis of LSU sequences of both taxa from Indonesia has also shown differences, because a partial sequence of the large subunit of the rRNA gene (LSU; length 1438 bp) differed in 0.974% (sequence similarity 0.990) (J. Brabec – unpublished data).

The taxa redescribed above may occur sympatrically and may infect even the same host specimen of *Saurida tumbil* and *S. undosquamis*. On the other hand, *S. longimanus* and probably *S. nebulosa* harbour only *O. sauridae*, whereas *S. micropectoralis* is infected with *P. ganapattii* only (Rao 1954, 1960; Subhadrappa 1955; Devi 1975; Shinde 1975; Jadhav & Shinde 1981; Khalil & Abu-Hakima 1985; Kuchta *et al.* 2008b; present study). Kuchta *et al.* (2008b) put wrongly *S. micropectoralis* as a host of *O. sauridae*. Neither *O. sauridae* nor *P. ganapattii* were found in *Saurida gracilis* (Quoy & Gaimard) from New Caledonia. *O. sauridae* is more widely distributed in the Indopacific and occurs from the Persian Gulf and coast of Mozambique throughout India and Indonesia up to the Pacific coast of Australia, New Caledonia and Japan. On the other hand, *P. ganapattii* was found only in the coastal waters of India and Indonesia, but never in the Pacific Ocean (Subhadrappa 1955; Devi 1975; Radhakrishnan *et al.* 1983; present study).

Radhakrishnan *et al.* (1983) observed seasonal variation in the prevalence of infection of *S. tumbil* off India, with the highest prevalence (up to 100% for *P. ganapattii* and up to 20% for *O. sauridae*) from October to November and the lowest from January to February (35%). The present values from *S. tumbil* are much lower (9% in *O. sauridae* and 4% in *P. ganapattii*), but most hosts from Indonesia were dissected in the rainy season (from January to March) when the prevalence is probably at its minimum (Radhakrishnan *et al.* 1983).

Several authors described the cysts with the scolex of *P. ganapattii* occurring in the fish liver (Rao 1954; Subhadrappa 1955; Radhakrishnan *et al.* 1983), but this site of infection was not confirmed in the newly collected material. In fact, the tapeworm forms cysts in the wall of the pyloric caeca, immediately next to the liver, which may have given the impression of its location in this organ. Penetration through the intestinal wall and the presence of a *scolex deformatus* is known also in another bothriocephalid cestode, *Anantrum histiocephalum* Jensen & Heckmann, 1977, and two species of triaenophorids, namely *Abothrium gadi* Beneden, 1871 and *Parabothrium bulbiferum* Nybelin, 1922, but none of these species form extraintestinal cysts (Kuchta *et al.* 2008b).

Palm (2000, 2004) reported 54 different trypanorhynch cestodes species from the southern Java coast, Indonesia. Lönnberg (1893) and Yamaguti (1954) listed 5 elasmobranch cestodes from Java and Sulawesi, and recent studies describe further tetraphyllidean cestodes from Borneo (Caira *et al.* 2007; Twohig *et al.* 2008). On the contrary, only four bothriocephalideans have been reported so far from Indonesia, namely *Anchistrocephalus aluterae* (Linton, 1889), *Oncodiscus sauridae*, *Paraechinophallus* sp., and *Penetrocephalus ganapattii* (Kuchta *et al.* 2008b), making this region of major scientific interest for future fish parasitological research.

Finding *O. sauridae* in New Caledonia represents the first bothriocephalid tapeworm reported from these waters (Justine 2007).

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