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# *Carinotetraodon imitator*, a new freshwater pufferfish from India (Teleostei: Tetraodontiformes)

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## Abstract

*Carinotetraodon imitator*, new species, a sexually dimorphic freshwater pufferfish, is described from Kerala, India. It has a colour pattern resembling that of *Tetraodon travancoricus*. The two species differ, however, in details of the colour pattern, in body spination, and the presence of an erectile dorsal and ventral keel of skin in courting males; the latter character assigns the new species to the genus *Carinotetraodon*. It is distinguished from *C. lorteti* and *C. salivator*, by colour pattern of both sexes, body spination, and a maximum size of less than 26 mm standard length which makes *C. imitator* one of the smallest known pufferfishes. The keels have not been observed in *T. travancoricus* but this species is hypothesized to belong to *Carinotetraodon* on the basis of osteological characters.

Key words: Tetraodontidae, *Carinotetraodon*, sexual dimorphism, skin keels

## Introduction

Hora & Nair (1941) described the pufferfish *Tetraodon travancoricus* from the Pamba River in Travancore (now Kerala State), southwestern India. The species is noteworthy for its small maximum known size of less than 25 mm standard length, making it the smallest known member of the family Tetraodontidae. Since its description, only little information on this species has been available. In his revision of Asian freshwater tetraodontids, Dekkers (1975) was not able to examine material of *T. travancoricus* and could only repeat Hora & Nair's (1941) information. Dekkers (1975) classified *T. travancoricus* along with *T. cutcutia* and *T. lorteti* in a *cutcutia*-group characterized by undivided nasal tubes, whereas the nasal tubes are distally divided into two lobes in the other Asian freshwater puffers. Åhlander (1988) caught *T. travancoricus* near Kottayam, Kerala, and provided some additional

information on this poorly known species. Talwar & Jhingran (1992) considered it as 'probably one of the endangered species', without providing arguments to support this assumption. Recently, Inasu (1993) commented on the sexual dimorphism of *T. travancoricus*, of which he had caught a 'good number' at Pudukkad, Trichur, Kerala.

In 1992, R. Pethiyagoda and the second author found *T. travancoricus* to be locally abundant in some coastal lagoons of Kerala. The species has been a recent feature of aquarium fish exports from India and most specimens are reportedly caught in the vicinity of Cochin, Kerala. Among the exported fish, we observed that some specimens had a slightly different colour pattern. Closer examination demonstrated that they represent a second species of dwarf puffer differing from *T. travancoricus* in several important aspects

of coloration and body spination. Males of the new species erect conspicuous middorsal and midventral keels of skin during courtship; this is the main character which Benl (1957) used to diagnose *Carinotetraodon*; such keels are not known in species of *Tetraodon*. Dekkers (1975) considered *Carinotetraodon* a synonym of *Tetraodon*, but Tyler (1978, 1980) treated it as a valid genus on the basis of its osteology and external features. This opinion was followed by Kottelat et al. (1993) and Lim & Kottelat (1995). Thus, the new species is described here as *Carinotetraodon imitator*.

The new species is known only from specimens obtained through the aquarium trade. Despite our efforts we have not been able to obtain material with more detailed and reliable locality data. The decision to publish the description of a new species without detailed locality data may appear anachronistic, but we feel that under the present circumstances it is justified. With or without locality data, the species exists and needs a name so that we can report observations on morphology and reproduction. To postpone a formal naming of the species until samples with reliable locality information become available would mean postponing publication of our observations by several years and we do not think that this is advisable. Considering that the species has already been mentioned and illustrated several times in the aquarium literature, delays increase the risk of a name being 'accidentally' made available for it; experience shows that the problems resulting from 'accidental' creation of new names is a nuisance much more significant than the enigma of the precise origin of the material. We also believe that formally naming the species is the best way to call attention to its existence and will incite others to look for it.

#### Material and methods

Measurements were made with electronic callipers following the method of Dekkers (1975) and Lim & Kottelat (1995). Terminology for the dark marks on the body is explained in Figs. 5 and 6. The examined material is preserved in the following collections: AMNH, American Museum of Natural History, New York; CMK, collection of second author; NRM, Swedish Natural History Museum, Stockholm; ZRC, Zoological Reference Collection, Singapore. SL stands for standard length and HL for head length.

Clearing and staining follows the method of Dingerkus & Uhler (1977), preparation of eggs and larvae for SEM that of Britz et al. (1995), and PAS (periodic acid Schiff) staining to demonstrate adhesive cells that of Peters & Berns (1982).

#### *Carinotetraodon imitator*, new species

(Figures 1-3)

**Holotype.** ZRC 42593, male, 25.6 mm SL; aquarium specimen, reportedly from India: Kerala: Cochin District, in small rivers; imported on 20.01.98, preserved 03.02.98, Aquarium Glaser, Frankfurt.

**Paratypes.** AMNH 211073, 1 male, 21.3 mm SL, 1 female, 19 mm SL, same data as holotype; ZRC 42594, 1 female 21.1 mm SL, same data as holotype; NRM 42003, 1 male (26.4 mm SL), 1 female (20.7 mm SL), aquarium specimens, same data as holotype, but imported Nov. 96; CMK 14810, 1 female (21 mm SL) with large ovaries, aquarium specimen, same data as holotype, but imported Nov. 96; AMNH 211074, 1 male (26.5 mm SL), 1 female (20.5 mm SL) c&s, aquarium specimens, same data as holotype, but imported Nov. 96.

#### Diagnosis

*Carinotetraodon imitator* is a small, sexually dimorphic pufferfish, fully mature already at slightly over 20 mm SL. It is distinguished from the other species of *Carinotetraodon* by the following unique characters: both sexes (preserved specimens) with a pattern of black blotches, females with numerous additional tiny spots (see Figs. 1-3) (vs. broad stripes in males and reticulated line pattern in females of the other two species); body spination (see Fig. 7) consisting of few, slender pointed spines (vs. densely covered with broad and blunt spines in *C. lorteti* and in *C. salivator*); more pectoral-fin rays (17-18, vs. 16-17 in *C. salivator* and 14-15 in *C. lorteti*); fewer dorsal- (9-10) and anal-fin rays (8-9) (vs. D 10-11, A 10 in *C. salivator* and D 11-12, A 10-11 in *C. lorteti*).

The colour pattern of *C. imitator* is similar to that of *T. travancoricus* (compare Figs. 3 with 4 and 5 with 6) but the species can be easily distinguished from *T. travancoricus* by the following characters: body spination (see Fig. 7): few slender pointed spines (vs. dense coverage with slender, pointed spines), males with pale spots (vs. distinct blotches), females with numerous tiny black spots interspersed among larger blotches (vs. larger blotches, rarely with few tiny spots), courting males with erectile middorsal and midventral keels of skin (Fig. 3b) (vs. no keels).

#### Description

Measurements are from the holotype and three paratypes (19.0-25.6 mm SL): total length 126.6-130% SL, maximum body depth 37.4-40.9 % SL, maximum body width 32.0-36.5 % SL, length of caudal peduncle 17.1-20.7% SL.

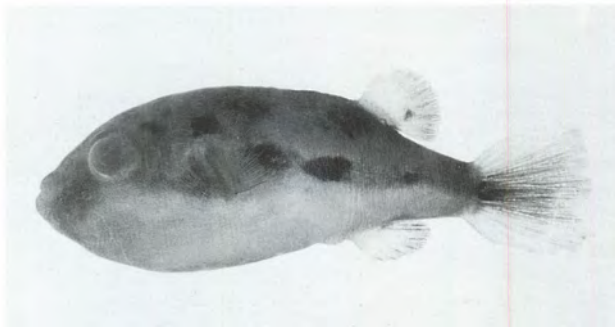


Figure 1. *Carinotetraodon imitator*, holotype, male, 25.6 mm SL.

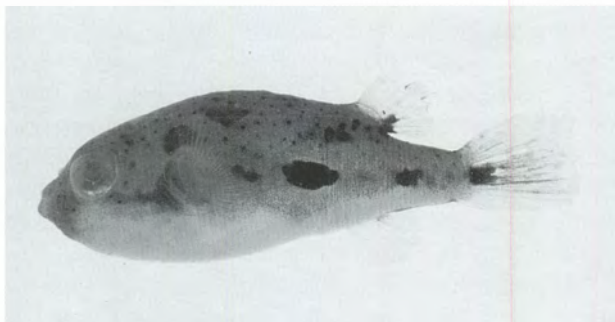


Figure 2. *Carinotetraodon imitator*, one of the paratypes, female, 21.1 mm SL.

Head length 39.0–41.8% SL, eye diameter 12.1–14.2% SL, 29.5–36.5% HL. Mouth terminal, directed slightly upward and lower lip protruding slightly beyond upper lip; chin heavy in males, more so than in females; mouth opening below level of eye. Nostrials short and tubular, closer to anterior end of eye than to mouth, with two short opposing lobes at the rims inclined toward each other. Dorsal-fin rays 9 (8) or 10 (1), anal-fin rays 8 (3) or 9 (6), pectoral-fin rays 17 (4) or 18 (5), caudal-fin rays 5+6 (9). Body spination consisting of few slender pointed spines on the anterior body (Fig. 7a).

**Coloration.** Male, preserved (Fig. 1). Head and body dirty yellow to greyish brown. A pale grey band from lower lip below eye to gill opening. Pale supraorbital blotches (for terminology of dark marks see Fig. 5). Two suprapectoral blotches above pectoral fin, the anterior one dark and distinct, the posterior one paler, located above and slightly behind the anterior one. A darker dorsal-fin blotch close to anterior end of dorsal fin. Three defined dark blotches on the flanks of the body: a postpectoral blotch posterior to pectoral fin, a conspicuous lateral blotch at level of anterior end of dorsal fin, and a peduncular blotch at level of posterior end of dorsal fin on the caudal peduncle. A dark mark at the base of caudal fin, the caudal blotch, extends as a broadening area to the end of caudal fin,

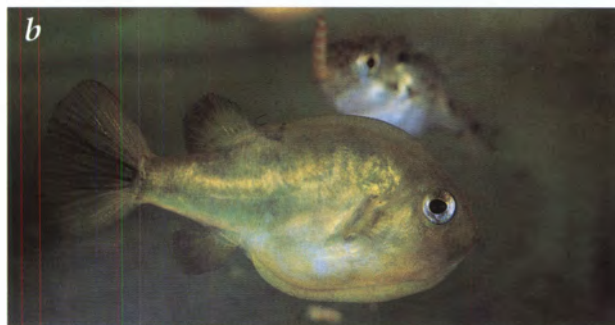


Figure 3. *Carinotetraodon imitator*. (a) live male in normal coloration; (b) in courting coloration with skin keels erected; (c) live female in normal coloration.

with upper- and lower-most margins of caudal fin yellowish. Depending on the individual these dark body marks may be conspicuous or faint or be broken up into several smaller dark spots. Pectoral, dorsal and anal fins dirty yellow becoming brownish distally. Ventrums dirty white to dirty yellow, becoming brownish toward midventral line and forming a faint stripe from lower lip to caudal peduncle surrounding anal fin where stripe is broader and darker. In life (Fig. 3a–b), body including ventrum and fins dirty yellow to bright yellow with the darker marks being paler but still visible. Iris blueish. Sexually active males (with erected dorsal and ventral keels) bright yellow without darker marks. Only middle of caudal fin and distal areas of dorsal and anal fins brownish.

Female, preserved (Fig. 2). Head and body light brown to beige with white ventrum. Dark band from lower lip below eye to gill opening as in males. Nu-

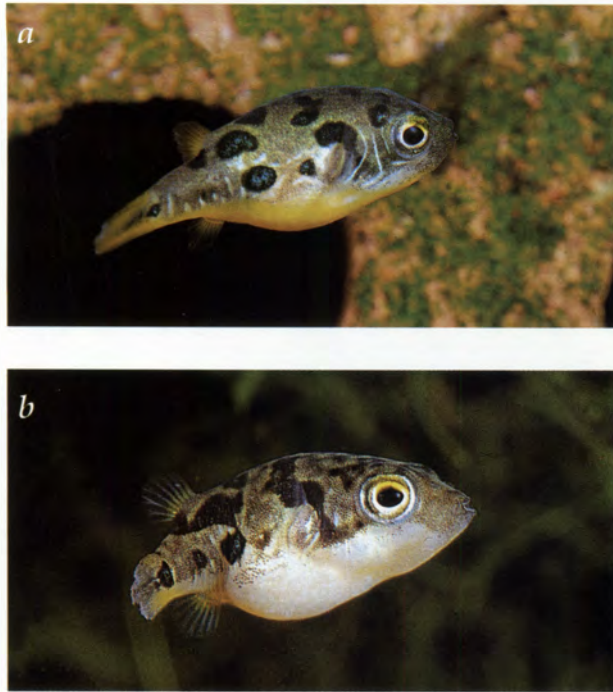


Figure 4. *Carinotetraodon travancoricus*, live coloration of (a) male; (b) female.

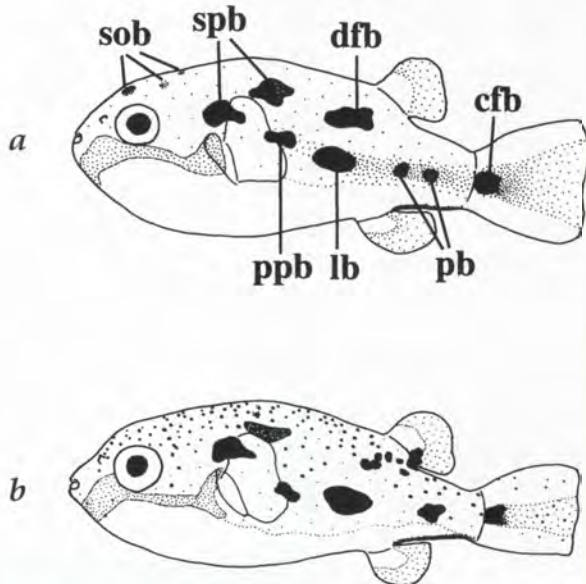


Figure 5. *Carinotetraodon imitator*, schematic drawing of colour pattern in (a) male; (b) female. Abbreviations: cfb : caudal-fin blotch, dfb : dorsal-fin blotch, lb : lateral blotch, pb : peduncular blotch, ppb : postpectoral blotch, sob : supraorbital blotch, spb : suprapectoral blotch.

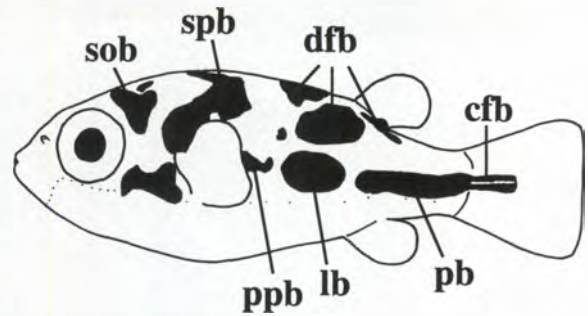


Figure 6. *Carinotetraodon travancoricus*, schematic drawing of colour pattern, after drawing of holotype from Hora & Nair (1941), for abbreviations see Fig. 5.

merous tiny black spots all over dorsal and lateral parts of head and body, absent from ventrum. Suprapectoral blotches present, anterior one dark, posterior one either pale or dark depending on the individual. Dorsal-fin blotch small and pale, often broken up into several small spots. Darker spot at posterior base of dorsal fin extending into fin in one female. Row of three lateral spots present but only lateral blotch conspicuous, postpectoral and peduncular blotch sometimes broken up into several small spots. Caudal-fin blotch well developed.

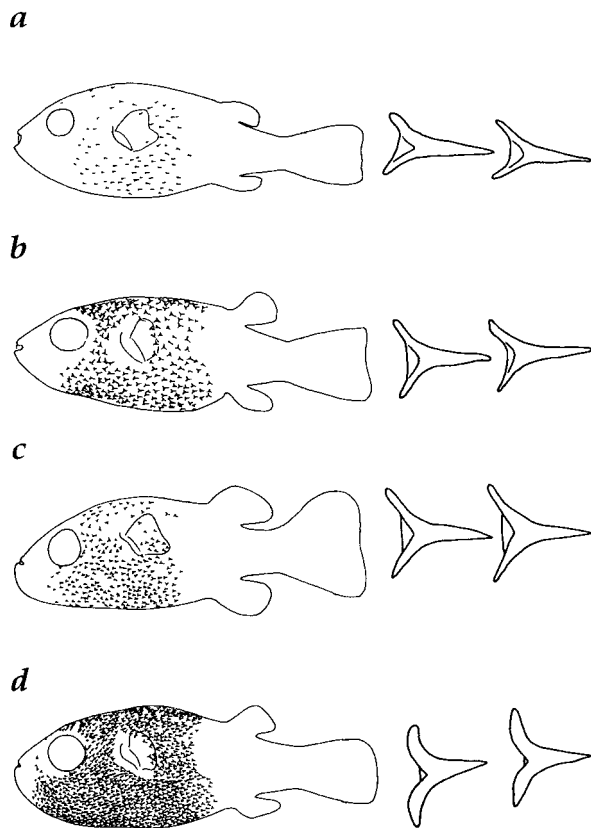
As in males, depending on the individual these dark body marks may be conspicuous or faint or be broken up into several smaller spots in females too. Caudal fin translucent and brownish-beige, with several tiny black spots as on body and with middle part darker than upper and lower margins. Pectoral, dorsal, and anal fins brownish translucent. Midventral brown stripe on ventrum lacking in females, area around anal fin darker, sometimes extending as short stripe to lower half of caudal-fin base. In life, coloration similar to that of preserved specimens but large and tiny spots more pronounced (Fig. 3c). In the same individual, large blotches can be conspicuous and entire at one time and at another time be broken up into several smaller spots thus appearing faded.

**Distribution.** According to the information obtained from the exporter through the importer, *C. imitator* occurs in small rivers in the Cochin District, Kerala, India.

**Etymology.** The name *imitator* (from the Latin, with same meaning as in English) alludes to the colour pattern which closely resembles that of *Tetraodon travancoricus*. A noun in apposition.

#### **Ethology, egg and larval structure**

Five specimens of *C. imitator*, two males and three females, were kept in a 70 l tank. The water temperature varied between 20–24°C. The water was slightly



**Figure 7.** Schematic representation of body spination and individual spines from area below lower pectoral fin base. (a) *Carinotetraodon imitator*, (b) *C. travancoricus*, (c) *C. lorteti*, (d) *C. salivator*.

brackish (10 l of sea water per 60 l of freshwater). After a few weeks, males started courting and soon thereafter the first spawnings could be observed. Courting males erected the dorsal and ventral keels while courting females or threatening other males. Males tried to attract females to the spawning site by swimming back and forth between spawning site and females. In addition they tried to direct females by actively pushing them with their body. If a female was ready to spawn she followed the leading male. Spawning occurred among branches of Java moss (*Vesicularia*). Before eggs were released, she pressed close to his body, both curved to a half circle. Then one or two eggs were released during the spawning bout. Up to ten or more spawning bouts per spawning sequence, which lasts a few hours may follow. However, the number of eggs recovered from any one spawning sequence was never higher than 10. This may be explained by the other conspecifics following the spawning pair and frequently succeeding in eating the just-laid eggs even though the male defended

the spawning site. There was no fanning or other form of parental care except the defense of the spawning territory. This, however, may last several weeks because, once a spawning site is established, males prefer to keep on spawning at this site. Eggs were large, about 1.6 mm in diameter (Fig. 8a), compared to the female's size (21 mm SL). They seem to possess no obvious stickiness because they fall out of the moss if it is lifted and gently shaken. Eggs are spherical, with a micropyle at the tip of a wart-like protrusion (Fig. 8b-d). Eggs hatch after 7 days (at 22-24°C). Hatched larvae are 3.5 mm long with brownish body and two translucent vertical bands, one behind large yolk sac and one at end of caudal fin (Fig. 8e-g). Larvae are capable of attaching to substrates with the ventral side of their yolk sac. SEM along with PAS-staining showed that there are numerous single adhesive cells concentrated on the ventral yolk sac (Fig. 8d-f).

### Discussion

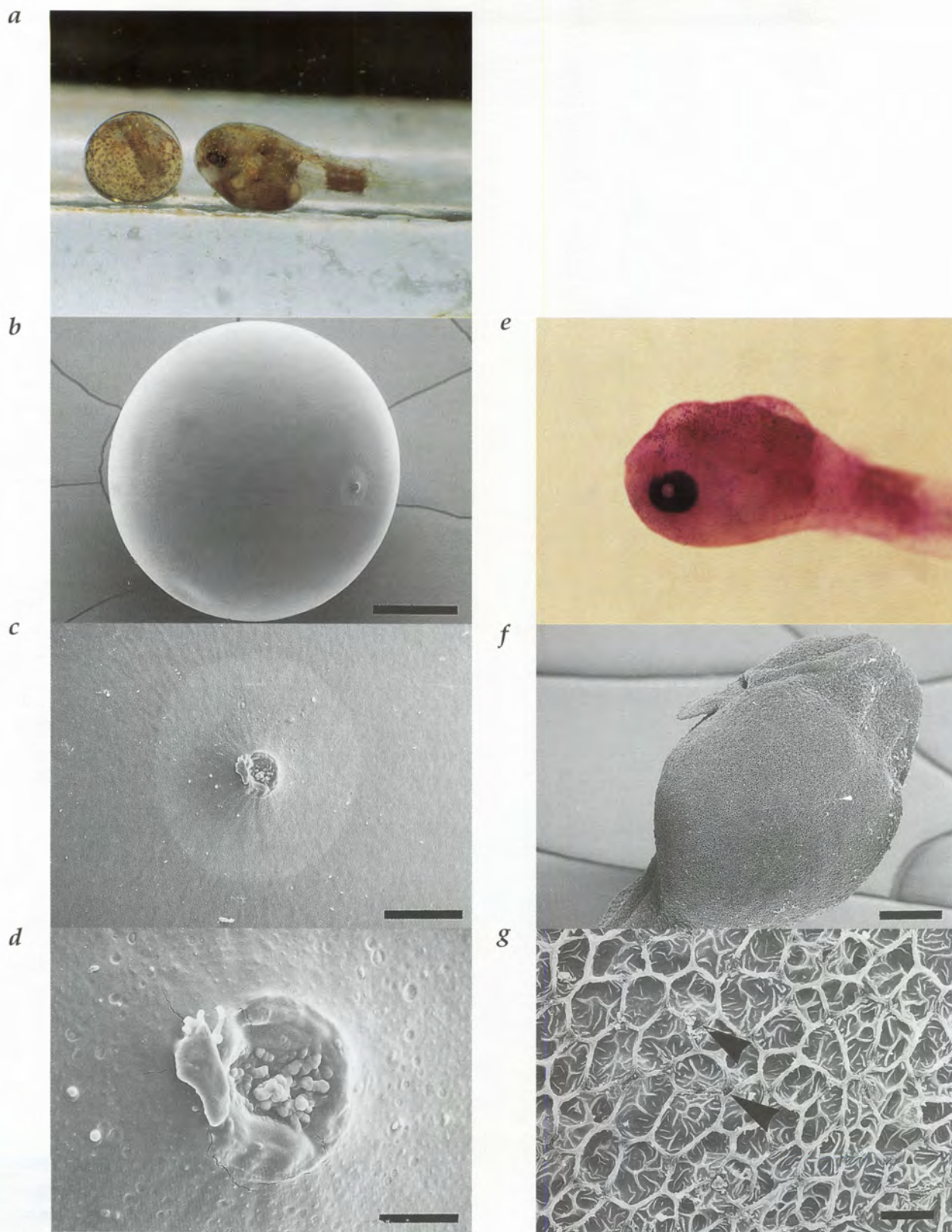
*Carinotetraodon imitator* is easily distinguished from its two congeners by its coloration in both males and females (Fig. 2). The body of male *C. imitator* is normally a dirty yellow to orange with several indistinct dark marks but becomes bright yellow to orange without any marks during the sexually active period. Males of *C. lorteti* and *C. salivator* never show this yellow colour and have a different colour pattern consisting of several broad light grey bands (Benl 1957, Tyler 1978, Lim & Kottelat 1995); in addition, male *C. salivator* uniquely have a mottled and reticulated pattern along the middle of the side and a variable number of dark brown bars on chin and on the sides, the latter possibly only present in the larger adult specimens.

Females of *C. imitator* differ from those of the other two species of *Carinotetraodon* in color pattern, too. They possess few large black blotches with numerous tiny black spots interspersed (Fig. 2). Females of *C. lorteti* and *C. salivator* have a pattern of reticulate lines along the body (Tyler 1978, Lim & Kottelat 1995) and no larger blotches or tiny spots.

Independent of the gender, *C. imitator* can be differentiated from its congeners by the lower count of dorsal- (9, rarely 10) and anal-fin rays (8-9) (vs. D 11-13, A 10-12 in *C. lorteti* after Dekkers [1975], D 10-11, A 10 in *C. salivator* after Lim & Kottelat [1995]) and the few spines on the body (vs. dense spination).

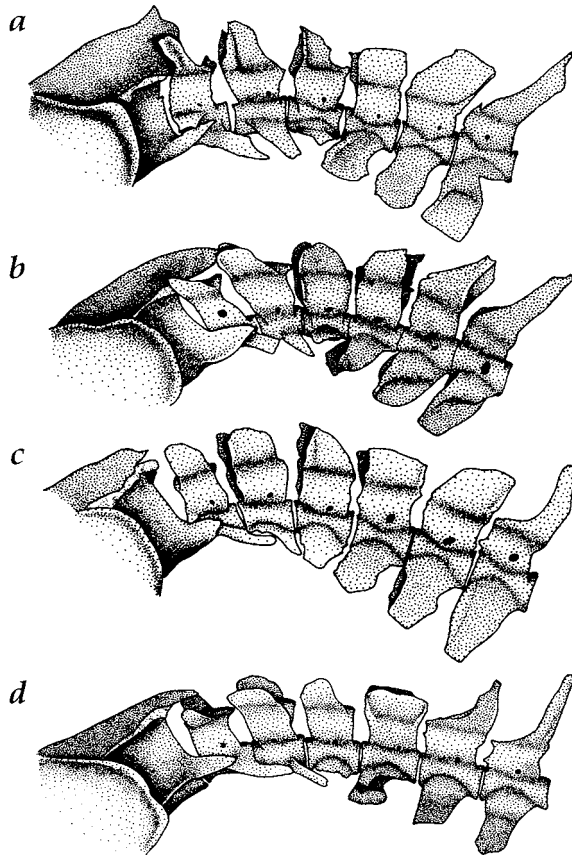
*Carinotetraodon imitator* also seems to be a smaller species (19.0-25.6 mm SL) than *C. lorteti* (up to 54 mm SL; Dekkers 1975) or *C. salivator* (up to 40 mm SL; Lim & Kottelat 1995).

Eggs and larvae also seem to distinguish *C. imitator* from *C. lorteti*. Richter (1983) reports spawned eggs of *C. lorteti* to be about 0.5 mm in diameter and very



**Figure 8.** *Carinotetraodon imitator*. (a) egg and just hatched, larva; (b) SEM-photograph of egg, with (c) detail of micropylar area, and (d) close-up of micropyle; (e) PAS-stained larva showing adhesive cells concentrated on yolk

sac; (f) SEM-photograph of ventral yolk sac, with (g) openings of adhesive cells between epidermal cells, marked by arrowheads (scale bars: 400  $\mu\text{m}$  in b, 40  $\mu\text{m}$  in c, 10  $\mu\text{m}$  in d, 200  $\mu\text{m}$  in f, and 10  $\mu\text{m}$  in g).



**Figure 9.** Lateral view of posterior part of skull and six anterior abdominal vertebrae of (a) *Carinotetraodon imitator*, (b) *C. travancoricus*, (c) *C. lorteti*, (d) *C. salivator*. Note conspicuous ventrally directed parapophyses on vertebrae.

sticky. They are about 1.6 mm in *C. imitator* and have no obvious adhesiveness. Larvae of *C. lorteti* hatched after 60-72 hours and then measured 2 mm, whereas those of *C. imitator* hatched after 7 days and were 3.5 mm long. Unfortunately, there is no information on the reproductive behavior of *C. salivator*.

*Carinotetraodon imitator* has a sexually dimorphic coloration which resembles that of *T. travancoricus* in its basic pattern but otherwise is unique among freshwater puffers. It can be distinguished from *T. travancoricus* by the number and shape of body spines (few and shorter spines vs. numerous, longer and more pointed spines). Further characters distinguishing it from *T. travancoricus* are the faded dark spots in males (vs. distinct spots) and the numerous tiny black spots of females (vs. no or rarely few tiny spots).

Nevertheless, it is amazing how the basic pattern of the larger black blotches in *T. travancoricus* resembles the pattern of the dark black spots and marks in *C. imitator*. In *T. travancoricus* (Figs. 4, 6) there are usually supraorbital blotches, suprapectoral blotches

which may be confluent or separate, a postpectoral blotch, a lateral blotch, a group of dorsal-fin blotches, the peduncular blotch usually present as a band on the caudal peduncle, and a caudal spot on the base of the caudal fin. Hora & Nair (1941) reported that coloration varies considerably in *T. travancoricus* but as evidenced by a larger sample (CMK 8644, n=87) variation only concerns size or number of these spots or connections between them but not their location on the body. The different dark marks of *C. imitator*, conspicuous in females and somewhat faded in males, are located on the body in positions identical to those of the dark blotches in *T. travancoricus* (compare Fig. 3 with 4 and Fig. 5 with 6).

All specimens of *C. imitator* known to us have been observed in shipments of aquarium fishes exported from Calcutta along with *T. travancoricus*. According to the exporters, all these puffers are collected from Cochin District in Kerala. In the absence of contrary evidence, we accept this information and tentatively hypothesize that the two species really occur in sympatry and syntopy. But we feel that it still cannot be excluded that the two species come from different areas, either adjacent within Cochin District or very distant within India. We also cannot exclude that stocks from different areas regularly get mixed by middlemen unaware of the existence of several species of dwarf puffers.

The two species apparently are the smallest puffers known to date. *Carinotetraodon imitator* is already sexually mature at sizes of slightly over 20 mm SL. Dekkers (1975) speculated that the type specimens of *T. travancoricus*, which measured 20.8-22.0 mm SL, were only juveniles and that the species would grow much larger. Åhländer (1988), however, demonstrated that females were already fully mature and had ripe eggs at a standard length of 18.8 mm.

The eggs measured 1.0 mm on average. With the normal size increase after spawning, which is due to the formation of the perivitelline space (Laale 1980), spawned *T. travancoricus* eggs may be slightly smaller than those of *C. imitator*.

#### Remarks on live coloration in *T. travancoricus*

For the description of *T. travancoricus*, Hora & Nair (1941) had examined five preserved specimens obtained from the Pamba River, Central Travancore. The color description is thus based on preserved material only. Concerning the live coloration, the authors cited the collector's note that 'The general yellow colour of the body has faded now, but when fresh it gives the fish a beautiful appearance'. Recently, Inasu (1993) reported sexual dimorphism in *T. travancoricus* and briefly commented on its live coloration.

With the availability of live specimens and their observation in captivity for several months, it is now possible to provide a more detailed description of their live coloration and supplement Inasu's data. We confirm that there is a sexual dichromatism. Males have a light brown background colour with a bright yellow ventrum. There are several black blotches on the body (Figs. 4, 6). There are several thin iridescent blue lines around the eye not mentioned and probably overlooked by Inasu (1993). They vary in number and almost completely fade away in preserved specimens in which only faint whitish lines can be discerned. Inasu (1993) reported males to have a 'large number of narrow dark lines on the caudal fin' which are lacking in females. Our live and preserved specimens do not confirm any sex-related striking differences in coloration of the caudal fin (sex was checked by dissection). Also, contrary to Inasu (1993), our observations do not confirm that differences in darkness of the body blotches allow one to distinguish the sexes reliably; their coloration is correlated with the mood of the individual fish. A 'dark bluish band from mouth to the caudal' persisting 'even in preserved specimens' also does not seem to be a reliable character to distinguish sexes because we found males without this band among the live and the preserved specimens. Inasu (1993) also mentioned that males can be distinguished from females by their size, with males growing larger than females, but without providing actual data supporting this statement. Our material does not support a sex-related size difference.

Females have a similar colour pattern but differ in having a darker brown ground colour and a white ventrum which may be yellowish in some specimens but never bright yellow as in males. This difference in ventrum coloration was already mentioned by Inasu (1993). In addition, females lack the iridescent lines around the eye which are present in males. In both sexes the iris has an outer brown ring and an inner one which is iridescent yellow.

#### Systematic position of *T. travancoricus*

Benl (1957) proposed the new genus *Carinotetraodon* for *C. lorteti* based on the ability of courting males to erect middorsal and midventral keels. Dekkers (1975, p. 93) did not accept *Carinotetraodon* as a valid genus and recommended to 'await the results of osteological studies before adopting this view'. Tyler (1980), investigating the osteology of *C. lorteti*, presented evidence for the validity of *Carinotetraodon*. He suggested that this genus may be the closest relative of *Canthigaster*, a marine puffer genus. One synapomorphy of these two genera is the presence of distinct parapophyses (basiventrals) on the first ab-

dominal vertebrae. The parapophyses on abdominal vertebrae 1-3 are paired and do not meet in the midline. Those on subsequent vertebrae meet in the midline to form a complete canal for the dorsal aorta and also possess flattened hemal spines. In *Canthigaster* these hemal spines have posterior lobes which are not present in *Carinotetraodon*. The distinct and modified parapophyses on the first abdominal vertebrae occur in *C. lorteti*, *C. salivator* and in *C. imitator* (Fig. 9).

A closer examination of the osteology of *T. travancoricus* reveals that this species too has the modified parapophyses of *Carinotetraodon* (Fig. 9b). It also shares with the three species of *Carinotetraodon* a reduced number of vertebrae (17-18), small size and the presence of sexual dimorphism. As a consequence of these results we transfer *T. travancoricus* from the genus *Tetraodon* into the genus *Carinotetraodon*. From the colour pattern it seems that on the one hand *C. lorteti* and *C. salivator* and on the other hand *C. travancoricus* and *C. imitator* may be sister species.

Future studies may show whether *C. travancoricus* also has the ability to erect middorsal and midventral keels during courtship. The two males that were maintained in an aquarium for several months together with two females never erected keels. If keel erection, however, is strictly linked with courting or spawning, then it seems possible that male *T. travancoricus* may possess such keels, because the males we maintained did not display any courting behavior over the months of observation.

#### Additional material examined

*C. travancoricus*: all from India: Kerala: NRM 12181, 21 ex., 9.9-19.2 mm SL; Kottayam district, Vembanad Lake. - NRM, 12197: 1 ex., 14.5 mm SL; Kottayam district, Vembanad Lake bought at fish landing at Kumarakom boat jetty. - NRM, 12224: 1 ex., 18.7 mm SL; Kottayam district, Meenachil River drainage, Meenachil River and adjacent canals, NW of Kottayam. - NRM 12239, 7 ex., 10.3-14.4 mm SL; Kottayam district, Vembanad Lake, Kumarakom, lake and canals. - CMK 8644, 87 ex., 12.7-26.4 mm SL, of which 5 ex. c&s.

*C. lorteti*: aquarium specimens: CMK 14420, 5 ex., 28.4-35.8 mm SL. - pers. c&s coll. of first author, 1 ex. c&s 27 mm SL.

*C. salivator*: CMK 8393, 1 ex. c&s 25 mm SL.

#### Acknowledgements

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for providing most of the specimens of *C. travancoricus*, to Wolfgang Maier for permission to maintain the live materials in the tanks of the Lehrstuhl für Spezielle Zoologie, Universität Tübingen, Aquarium Glaser for cooperation and import of the specimens, Frank Schäfer for information on the collection area, Till Luckenbach for the colour photos, Martina Hohloch and Horst Schoppmann for technical assistance, and Thomas Stach for critical remarks on the manuscript and translation of Swedish literature.

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