Zool. Anz. **224** (1990) 1/2, S. 106—112
VEB Gustav Fischer Verlag Jena

On a New Trematode Belonging to the Genus *Velasquezotrema* Eduardo & Javellana, 1987 from *Bos indicus* in Tripura (India)

By Bishnupada Roy and Veena Tandon[1]

North-Eastern Hill University (India)

With 13 Figures (Accepted: May 17, 1989)

Key words: *Velasquezotrema*, new species, trematode, paramphistome, India

Abstract

Velasquezotrema tripurensis sp. nov. (Paramphistomidae) occurring in the rumen of *Bos indicus* L. in Tripura (India) is described. The new species is distinct from the only other known species of the genus, i.e. *Velasquezotrema brevisaccus* (Eduardo, 1981) Eduardo Javellana, 1987 in the presence of oesophageal sphincter, a new type of terminal genitalium, and also in surface features of the oral and acetabular regions. On account of the limited extent of the ventral pouch in *Velasquezotrema*, a new subfamily Velasquezotrematinae is proposed for the genus.

Introduction

Of the paramphistomid trematodes parasitizing the ruminant hosts in India, the pouched species are commonly represented by the genera *Gastrothylax* Poirier, 1883; *Carmyerius* Stiles et Goldberger, 1910; *Fischoederius* Stiles et Goldberger, 1910 (Yamaguti 1971, Mehra 1980, Srivastava 1982) and rarely by the genus *Duttiella* Srivastava, Prasad et Maurya, 1980. However, a survey of helminth parasites of bovine mammals in Tripura, a north-eastern state of India, revealed the presence of some paramphistomid flukes which were found not belonging to any of these genera but representing an hitherto undescribed form. The same are described herein as a new species under the genus *Velasquezotrema* Eduardo et Javellana, 1987, the only other known species of which is reported from the Philippines.

Material and Techniques

Twenty-four specimens of the present material were collected from the rumen of two freshly slaughtered cows, *Bos indicus* L., at Dharmanagar (24°23′N—92°10′E) in Tripura, India. Of these, 20 specimens were processed for light microscopy; after fixation in Bouin or 10% formalin, 14 whole mounts were prepared by staining in Borax carmine or Mayer's carmalum; four series of sagittal and two of cross sections were prepared and stained with haematoxylin and eosin. Ink drawings were made to scale with the aid of a camera lucida.

For scanning electron microscopy, specimens were fixed in 10% cold buffered formaldehyde and dehydrated through acetone, treated with TMS (Tetramethyl silane) following Dey et al. (1989),

[1] To whom all correspondence be addressed.

metal coated and observed under a Jeol JSM — 35 CF under electron acceleration voltage of 10—15 Kv.

The description is based on the above number of mounted and sectioned specimens. At least 10 eggs from each specimen were measured.

Results

Velasquezotrema tripurensis sp. nov. (Figs. 1—13)

Measurements and Description

Body elongated with rounded anterior and posterior ends, slightly bent in ventral direction, 6.780—11.752 mm long, 2.576—3.616 mm in greatest width at level little below genital pore; ratio of body breadth to body length 1:2.14 to 1:3.96.

Acetabulum subterminal or ventral, external diameter 0.587—0.858 mm, ratio to body length 1:9.37 to 1:16.64, of gastrothylax type as described by Näsmark (1937) (Fig. 4), units of dorsal and ventral circular muscles on lateral sides as follows: DE, 37—46; DI, 17—22; VE, 32—42 and VI, 16—24.

Oral aperture with long tegumental papillae; pharynx 0.316—0.497 mm long, 0.361—0.452 mm in maximum breadth, ratio to body length 1:16.66 to 1:28.88 and to diameter of acetabulum 1:1.29 to 1:2.38, of calicophoron type as defined by Dinnik

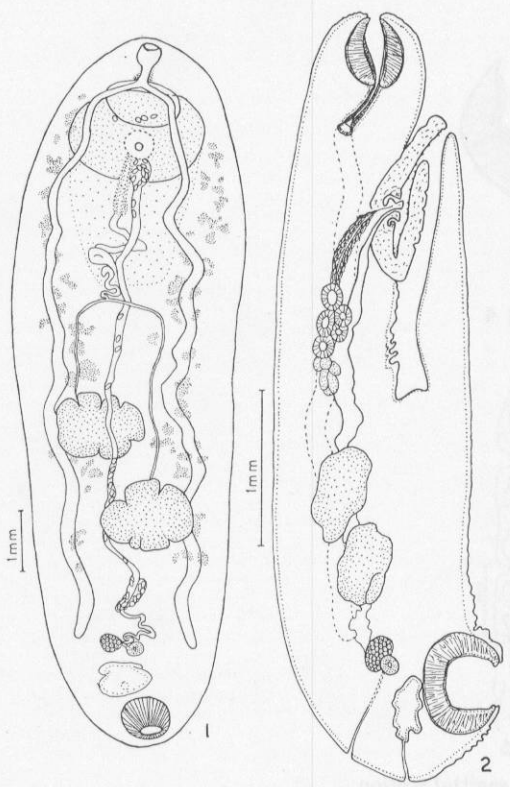
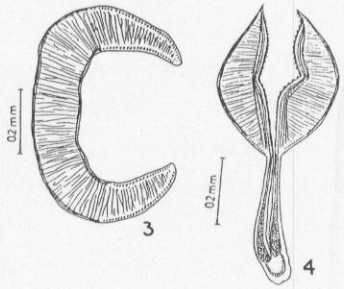


Fig. 1. Whole mount
Fig. 2. Whole worm, sagittal view. Note the extent of ventral pouch

(1964) in median sagittal section (Fig. 2). Oesophagus 0.180—0.271 mm long; ratio to body length 1:38 to 1:62.61; musculature of its wall thickened at its posterior end to form oesophageal sphincter, small gland cells extend along sides of pharynx and oesophagus and sometimes accumulate at posterior level of oesophagus. Caeca irregularly bent in dorsoventral direction, their blind end directed posteriorly or ventrally terminating at posterior level of ovary and Mehlis' gland.

Testes fourlobed, tandem or obliquely tandem, intercaecal or extended beyond caecal level in lateral direction, located in posterior half of body between posterior limit of ventral pouch and ovary and Mehlis' gland; anterior testis 0.994—1.265 mm long, 1.175—1.672 mm wide, posterior testis 0.813—1.220 mm long, 1.175—1.672 mm wide. Pars prostatica long, pars musculosa moderately developed, vesicula seminalis thin walled.

Ovary 0.226—0.361×0.180—0.452 mm, post-testicular; Mehlis' gland close to ovary; Laurer's canal not crossing excretory vesicle or duct and opening dorsally about 0.904—0.994 mm anterior (in sagittal section) to excretory pore. Uterus following median course with straight or sometimes with few loose coils in posterior part. Eggs 0.108—0.158 mm×0.054—0.090 mm. Vitellaria consisting of few loose or small clusters of vitelline glands in lateral fields or enchroaching into intercaecal regions and extending from little below oesophageal bifurcation to lower level of posterior testis or slightly beyond.



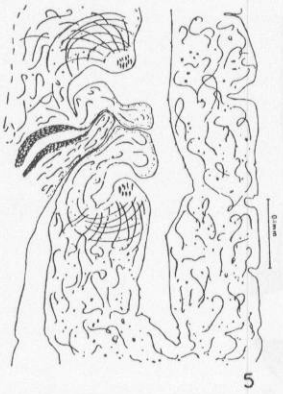


Fig. 3. Acetabulum, median sagittal section
Fig. 4. Pharynx and oesophageal sphincter, median sagittal section
Fig. 5. Terminal genitalium, median sagittal section

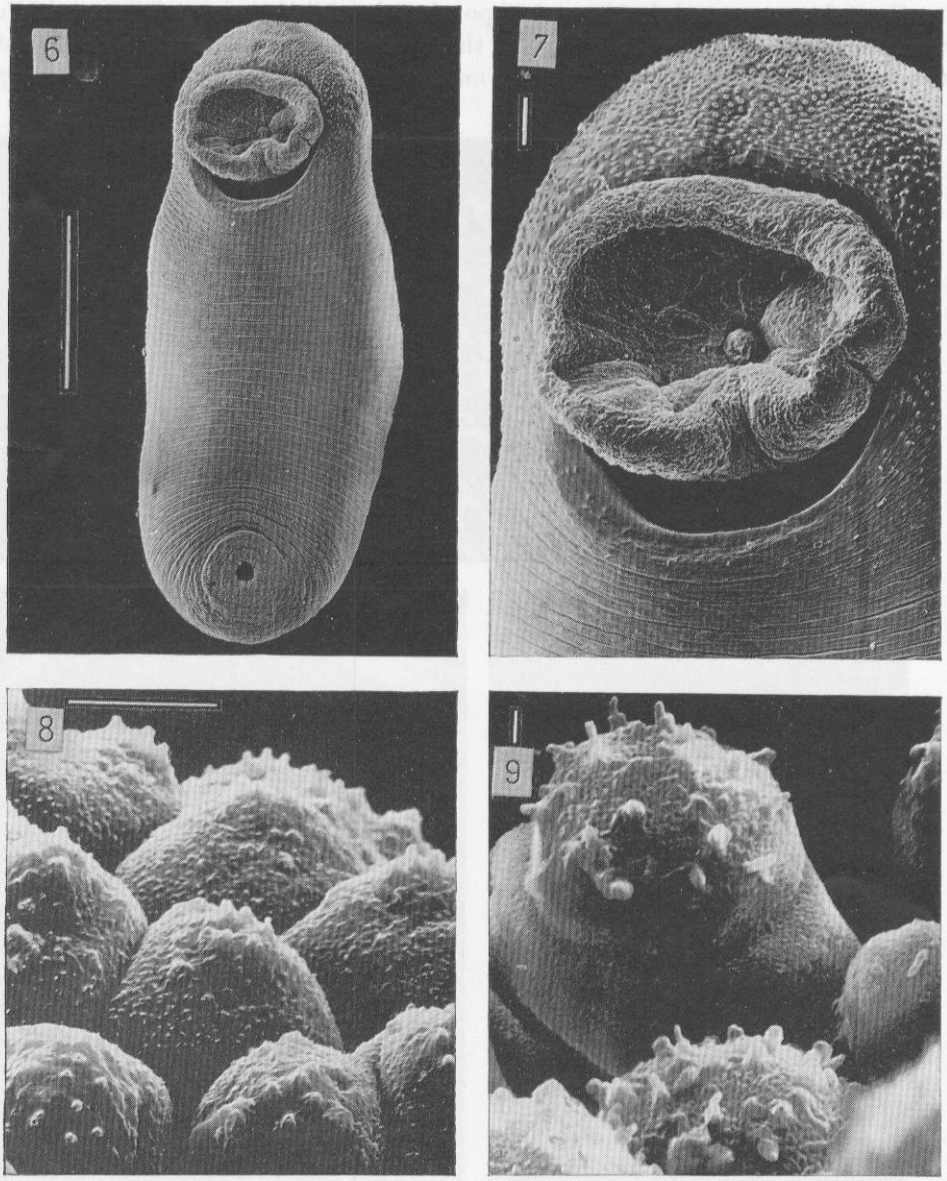


Fig. 6. Whole worm, ventral view. Note genital disc protruding out through ventral pouch opening (scale bar, 1 mm)

Fig. 7. Anterior end. Tegumental papillae around both oral and ventral pouch openings and genital papilla at the centre of genital disc (scale bar, 100 μm)

Fig. 8. Tegumental papillae around oral aperture in a magnified view (scale bar, 10 μm)

Fig. 9. A circum-oral papilla under higher resolution. Apical spine-like structure evident (scale bar, 1 μm)

Excretory vesicle between acetabulum and Laurer's canal, opening dorsally.

Genital pore ventral, inside ventral pouch at level little below oesophageal bifurcation. Terminal genitalium in middle of thick muscular and protrusible circular structure, which in extended condition comes out through ventral pouch opening and appears

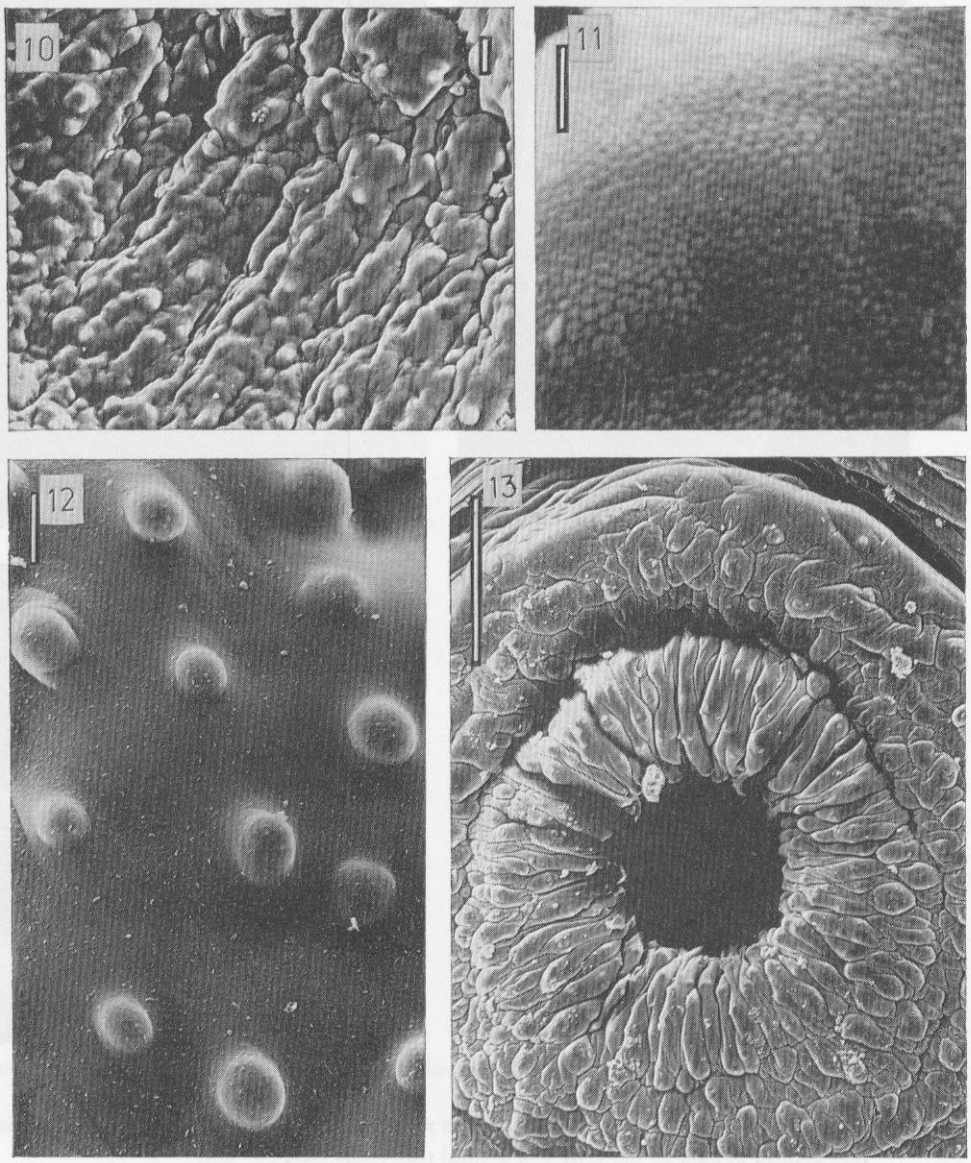


Fig. 10. Dome-shaped papillae scattered on the tegument of genital disc (scale bar, 10 μm)
Fig. 11. Enlarged view of a single papilla of genital disc. Note the finely tuberculated surface (scale bar, 1 μm)
Fig. 12. Tegumental papillae on general body surface (scale bar, 10 μm)
Fig. 13. Enlarged view of acetabular region (scale bar, 100 μm)

as a distinct circular disc on body surface (in w.m.); in withdrawn condition this muscular disc provides a deep extending ventral atrium, the inner walls of which are provided with wide-spaced and dome-shaped papillae in sagittal section; sphincter papillae weakly developed. This type of terminal genitalium is designated here as the tripurensis type (new type) (Fig. 3).

Ventral pouch narrow, more or less triangular, opening at level just above oesophageal bifurcation and extending posteriorly up to middle of body length, just anterior to anterior testis.

SEM examination of body surface (Figs. 6—13) revealed the presence of elevated dome-shaped papillae in the anterior one-fourth of the body. Under higher resolution, the circum-oral papillae are shown to possess several tentaculate or spine-like short processes at their apex. Similar papillate structures also surround the ventral pouch opening, more conspicuously on its lateral and anterior aspects; the region posterior to the ventral pouch opening lacks such papillae. Tegumental papillae also abound on the surface of the genital disc. The general tegument as well as the papillae exhibit a finely tuberculated appearance under high resolution.

Discussion

Eduardo and Javellana (1987) created the genus *Velasquezotrema* to accommodate *Fischoederius brevisaccus* Eduardo, 1981. They assigned it to the subfamily Gastrothylacinae of the family Gastrothylacidae along with other three genera, *Gastrothylax* Poirier, 1883; *Carmyerius* Stiles and Goldberger, 1910; and *Fischoederius* Stiles and Goldberger, 1910; and distinghished it from all of them on account of the limited extent of its ventral pouch which reaches only up to the middle of the body length. Yet another gastrothylacid paramphistome, *Duttiella cephaloporus* Srivastava, Prasad and Maurya, 1980 is characterized by a genital sucker which is present in no other genus of the family.

The present species also belongs to the genus *Velasquezotrema* owing to the presence of a small ventral pouch. However, it differs from the type species in several features: the greatest diameter of its body occurs in the region a little below the genital opening and not in the middle or posterior half of the body; its oesophagus is provided with an oesophageal sphincter; it possesses a terminal genitalium which is characterised by the presence of papillae in the wall of the protrusible ventral atrium, the presence of a genital sphincter and the absence of sphincter papillae. Besides, the presence of a circular and protrusible disc, which lodges the terminal genitalium, is a feature exclusive to the present form. This genital disc is very prominent and appears as mushroom-shaped in the live unfixed worms. In *V. brevisaccus* the terminal genitalium is lodged on a hemispherical bulge. In the present form the acetabular region lacks dome-shaped papillae and fine topography of the latter is also found to be different; the tuberculate apical structures of the papillae in the present form are not described for *V. brevisaccus*.

In view of these differences from *V. brevisaccus* the present form is considered a new species for which the name *V. tripurensis* is proposed.

Specific diagnosis

Oesophageal sphincter present, terminal genitalium tripurensis type i.e., presence of dome-shaped papillae in the wall of protrusible ventral atrium, sphincter papillae weakly developed.

On account of a ventral pouch with limited extent the genus *Velasquezotrema* is readily differentiable from other genera of the subfamily Gastrothylacinae in all of which the ventral pouch extends up to the posterior region of the body. In view of this, it seems justified to create a new subfamily Velasquezotrematinae to accommodate *Velasquezotrema*, now with two species under it.

Velasquezotrematinae n. subfamily

Diagnosis: Gastrothylacidae, with characters of the family. Ventral pouch limited in extent, extending posteriorly only up to middle of body.

Acknowledgements

The authors wish to thank Dr. S. L. Eduardo for his critical comments and suggestions. This study was supported by a research grant from the North-Eastern Council, Govt. of India to VT. Laboratory facilities provided by the Head, Department of Zoology, NEHU are thankfully acknowledged. We thank Dr. Sudip Dey of Regional Sophisticated Instrumentation Centre at NEHU for technical assistance in SEM study.

Deposition of specimens

Holotype (No. IV/ERS/ZSI/300), one paratype (No. IV/ERS/ZSI/301) and one series of sagittal and transverse sections each (No. IV/ERS/ZSI/302, 303) in Helminthological collection of Eastern Regional Station of Zoological Survey of India: Other paratypes and series of histological sections in Helminthological collection of the Department of Zoology, North-Eastern Hill University, Shillong (No. NEHU/Z-TM/1).

References

Dey, S., T. S. Basubaul, B. Roy, and D. Dey: A new rapid method using Tetramethyl Silane for preparation of soft biological tissue for scanning electron microscopy: Application to the trematode parasite *Homalogaster paloniae*, (1989), J. Microsc. (in press).

Dinnik, J. A.: *Paramphistomum sukumum* sp. nov. and other stomach flukes from cattle in the Sukumaland area of the Lake Region, Tanganyika. Parasitology **54** (1964) 201—209.

Eduardo, S. L.: *Fischoederius brevisaccus*, a new species of pouched amphistome (Gastrothylacidae) from ruminants in the Philippines. Syst. Parasit. **3** (1981) 1—6.

—, and C. R. H. Javellana: A new genus, *Velasquezotrema* for *Fischoederius brevisaccus* Eduardo, 1981. Philip. J. Vet. Med. **24** (1987) 29—34.

Maplestone, P. A.: A revision of the Amphistomata of mammals. Ann. Trop. Med. Parasit. **17** (1923) 113—205.

Näsmark, K. E.: Revision of the trematode family Paramphistomidae. Zool. Bidr. Upps. **16** (1937) 301—566.

Sey, O.: Revision of the family Gastrothylacidae Stiles and Goldberger, 1910 (Trematoda: Paramphistomata). Acta Zool. Hung. **29** (1983) 223—252.

Srivastava, H. D., A. Prasad, and A. C. Maurya: A new pouched amphistome *Duttiella cephaloporus* Gen. et sp., Nov. (Family-Gastrothylacidae) from buffaloes. Proc. Nat. Acad. Sci., India, **50** (1980) IV.

Stiles, C. W., and J. Goldberger: A study of the anatomy of *Watsonius* (n. g.) *watsoni* of man and of nineteen allied species of mammalian trematode worms of the super family Paramphistomoidea. Bull. hyg. Lab., U.S. Pub. Health and Mar. Hosp. Serv. 1981, No. 60, 259 pp.

Yamaguti, S.: Synopsis of digenetic trematodes of Vertebrates. Vols. I & II. Tokyo: Keigaku Publishing Co. 1971, 1074 pp.

Bishnupada Roy, Department of Zoology, North-Eastern Hill University, Shillong-793 014 (India)

Dr. Veena Tandon, Department of Zoology, North-Eastern Hill University, Shillong-793 014 (India)