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Surface Topography and Description of a Larval Didymozoid (Trematoda, Didymozoidae) from the Threadfin Bream *Nemipterus peronii*

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Abstract

An immature didymozoid larva is described from the marine fish *Nemipterus peronii* from the Arabian Gulf. The larva belongs to a group which is characterized by the presence of gland cells surrounding the distal part of esophagus and proximal parts of caeca. The topography of the tegument basically resembles that described for metacercariae of other digenetic trematodes. The tegumental surface is increased by the transverse ridges. On the ridges are protuberances which are prominent on the forebody and around the excretory pore. The domed papillae are found over the surface of the forebody particularly around the oral sucker and on the rim of the ventral sucker. The secretory pores occur around the oral opening and the excretory pore.

Key words: Didymozoidae, Kuwait, larval trematode, *Nemipterus peronii,* tegument, ultrastructure

Introduction

Adult trematodes of the family Didymozoidae (Poche, 1907) parasitize various predatory fishes, with the larvae found in small fishes and in a wide range of invertebrates (Nikolaeva, 1985). Yamaguti (1942), Fischthal and Kuntz (1964), Fischthal and Thomas (1968), Nikolaeva (1970), Kurochkin and Nikolaeva (1978) and Koie and Lester (1985) presented morphological descriptions of several new forms of didymozoid larvae found encapsulated in body muscles and mesentery or free in the digestive tract of numerous species of pelagic fishes. Studies using scanning electron microscopy on didymozoid larvae are not available.

The present study describes a larval didymozoid from body muscles of the marine fish *Nemipterus peronii* from the Arabian Gulf. Description of the larva is accompanied by scanning electron microscopy observations on the surface topography.

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Material and Methods

Living didymozoid larvae were isolated from the body muscles of infected threadfin bream, *Nemipterus peronii.* The fish was caught by gill netting in coastal waters of Kuwait in northern Arabian Gulf. The larvae were rinsed in 0.7% saline, fixed in FAA (formalin-acetic acid-alcohol) and stored in 70% ethanol. Whole-mounts were stained in acetic alum carmine or haematoxylin, cleared in xylol and mounted in Canada balsam. One larva was vitally stained in 0.5% neutral red and was drawn with the help of a Leitz drawing tube. Measurement of 10 whole-mounts were presented as means with ranges in parentheses and were given in micrometers.

Specimens for scanning electron microscopy were fixed in 2.5% glutaraldehyde in 0.05 M sodium cacodylate buffer pH 7.4 at 4°C for 3 h. The fixed specimens were then washed several times and post-fixed for 10 min in 1% osmium tetroxide in the same buffer, dehydrated in increasing concentrations of acetone and critical-point dried using CO_2 as transitional medium. The specimens were mounted on aluminium stubs, coated with gold/palladium and examined under a JEOL JSM-840 scanning electron microscope at an accelerating voltage of 15kV.

Results

Immature didymozoid larva
(Fig. 1)

Host : *Nemipterus peronii* (Valenciennes, 1830) (Pisces; Perciformes, Nemipteridae)

Habitat : Body muscles
Locality : The Arabian Gulf
Date : December, 1989
Specimens depository: The helminth collection of the Department of Zoology, University of Kuwait.

Description: Body 1974 (1378–2860) by 220 (182–260), elongate, subcylindrical, anterior part narrow, posterior extremity rounded. Forebody (anterior border of ventral sucker to anterior tip of body) 358 (224–449) its percentage of body length 18 (14–24). Ventral sucker 105 (90–108) in diameter, muscular. Oral sucker, 55 (48–65) by 40 (35–48), entirely muscular with terminal mouth. Pharynx 20 (15–25) by 16 (13–18), immediately posterior to oral sucker. Esophagus straight or slightly winding. Caecal bifurcation 76 (50–113)

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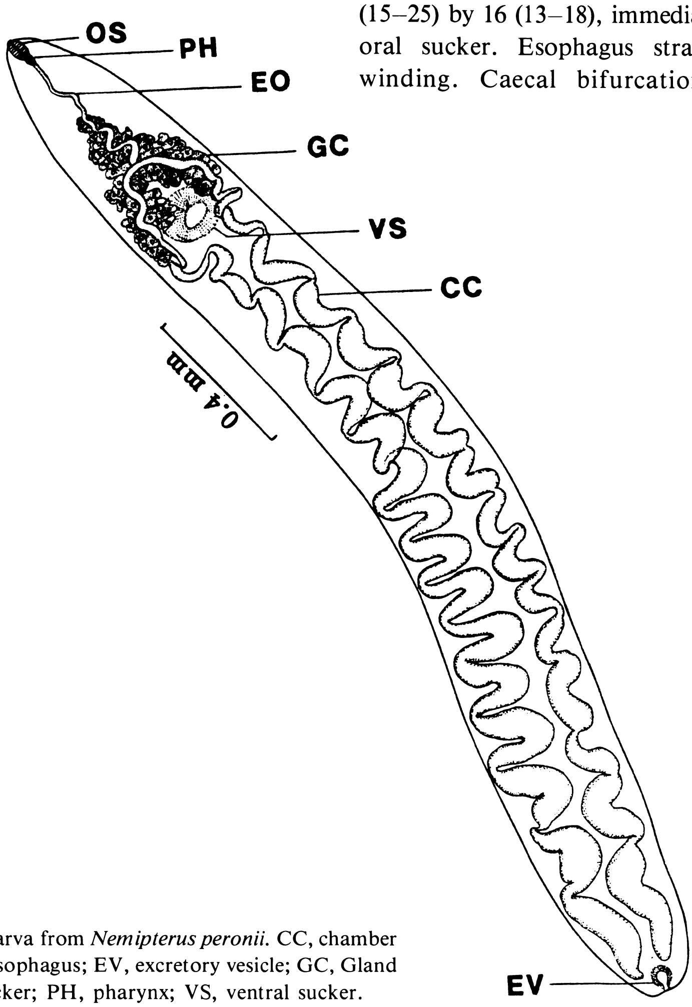


Fig. 1 Didymozoid larva from *Nemipterus peronii*. CC, chamber of caecum, EO, esophagus; EV, excretory vesicle; GC, Gland cell; OS, oral sucker; PH, pharynx; VS, ventral sucker.

anterior to ventral sucker. Caeca at ventral sucker level winding, continuing posteriorly into 15–16 U-shaped to oval chambers, sequentially becoming larger. Numerous gland cells surrounding distal part of esophagus and proximal parts of caeca. Excretory vesicle 53 (45–55) by 61 (52–85), postcaecal, pore terminal.

Surface topography: The body of the didymozoid larva could be divided into two regions based on the body shape and microtopography of the surface (Fig. 2): (1) a dorsoventrally flattened forebody extending from the oral sucker to the lateral aspects of the ventral sucker; and (2) a subcylindrical hindbody covering the area from the ventral sucker to the excretory pore at the posterior tip of the body. The body surface showed parallel transverse ridges roughly 0.5 μm broad (Fig. 3). At high magnification each ridge on the forebody appeared as a row of rounded microvilli 0.4 to 0.7 μm in diameter (Fig. 4). In contrast, the ridges on the hindbody appeared as smooth branching lamellae (Fig. 5). The ridges on the area around the excretory pore transformed to prominent finger-like microvilli (Figs. 6, 7). The areas around the oral sucker (Fig. 8) and on the rim of the ventral sucker (Fig. 9) were tightly packed with flat microvilli. The domed papillae with rough surfaces were present only on the forebody, particularly on areas immediately around the oral opening (Fig. 3) and the rim of the ventral sucker (Fig. 9). Few secretory pores were present in the vicinity of the oral sucker (Fig. 4) and the excretory pore (Fig. 7). Ciliated structures or spines are apparently absent.

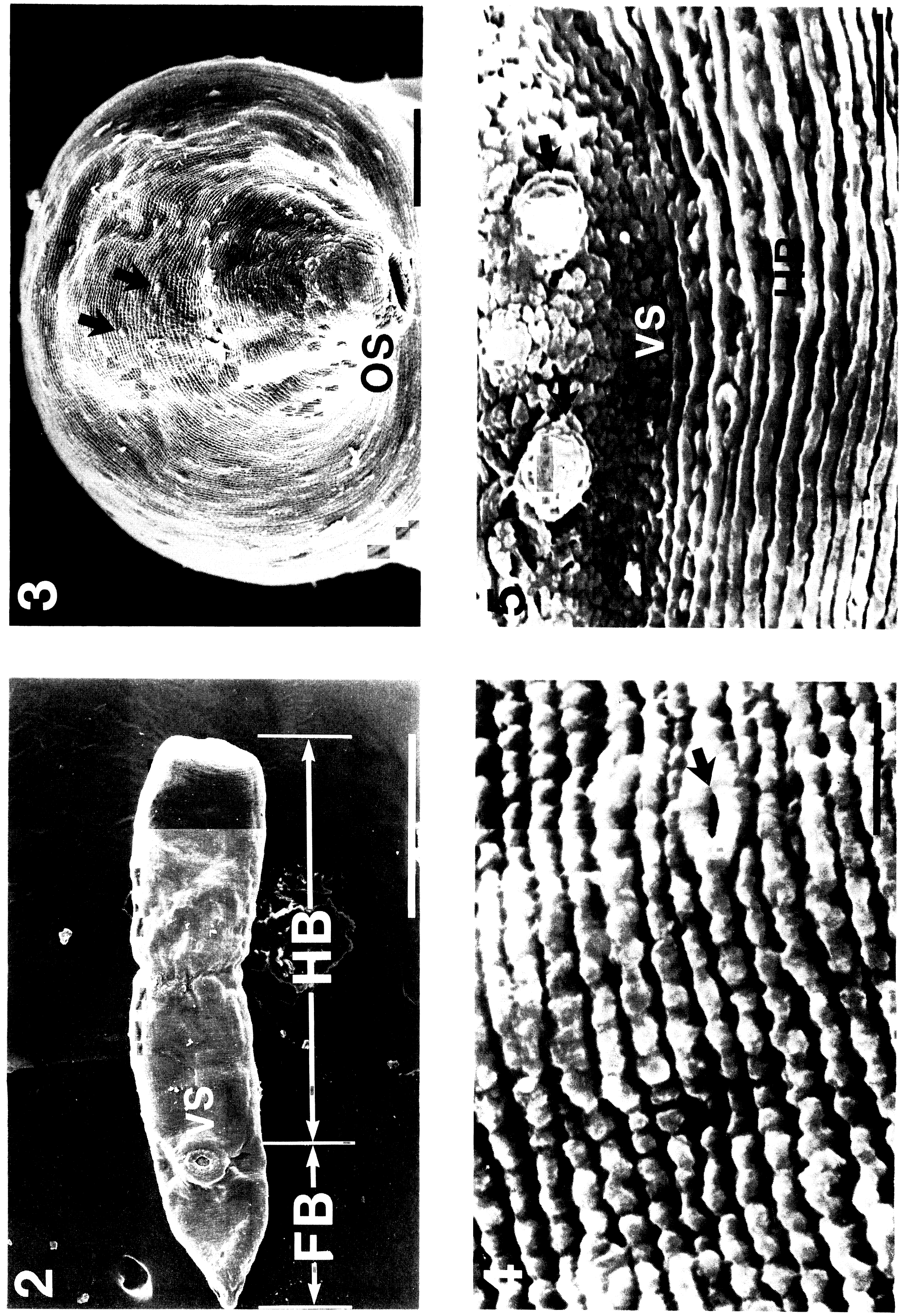
Discussion

Several species of marine fishes and invertebrates have been found harboring different forms of didymozoid larvae (Cable and Nahhas, 1962; Fischthal and Kuntz, 1964; Fischtal and Thomas, 1968; Madhavi, 1968; Reimer *et al.*, 1971, 1975; Overstreet and Hochberg, 1975; Yip, 1984; Koie and Lester, 1985). The life cycle of didymozoids is unknown, however, Nikolaeva (1965) speculated that Gastropod molluscs and planktonic invertebrates may act as the first and second intermediate hosts, respectively, while small fishes may serve as paratenic hosts. Attempts to classify didymozoid larvae (Nikolaeva, 1965; Yamaguti 1970, 1975; Kurochkin and Nikolaeva, 1978) have been largely unsuccessful due to unreliable structural criteria used in the proposed schemes. Therefore, designation of scientific names to didymozoid larvae have been discouraged in order to avoid forthcoming taxonomic confusion.

The didymozoid larva described in this study closely resembles immature "Didymozoid E" from small intestine of fishes from Cape Coast, Ghana (Fischtal and Thomas, 1968) and larval didymozoid "Species 5" from the body muscles and mesentery of fishes from Morton Bay, Australia (Koie and Lester, 1985). The principal feature which unite the three types of larvae is the presence of gland cells surrounding caecal bifurcation. It is relevant to note that similar glandular arrangement has been described in several species of mature didymozoids (Yamaguti, 1970, 1971).

Figs. 2–9 Scanning electron micrographs of didymozoid larva from *Nemipterus peronii.*

2. The ventral surface of entire specimen showing forebody (FB), hindbody (HB) and the ventral sucker (VS). Scale bar = 200 μm.
3. Anterodorsal view of forebody showing the oral sucker (OS), domed papillae (arrows) and ridges encircling the body. Scale bar = 20 μm.
4. High magnification of surface of forebody showing transverse rows of rounded microvilli and the secretory pore (arrow). Scale bar = 2 μm.
5. High magnification showing surfaces of the rim of the ventral sucker (VS) and hindbody (HB), and the domed papillae (arrows). Scale bar = 5 μm.
6. Tip of hindbody showing the microvilli around the excretory pore (EP). Scale bar = 20 μm.
7. High magnification of the body surface around the excretory pore showing the microvilli and secretory pore (arrow). Scale bar = 5 μm.
8. Anterior end showing oral sucker (OS) surrounded by domed papillae (arrows). Scale bar = 10 μm.
9. The ventral sucker (VS) showing distribution of domed papillae on the rim. Scale bar = 10 μm.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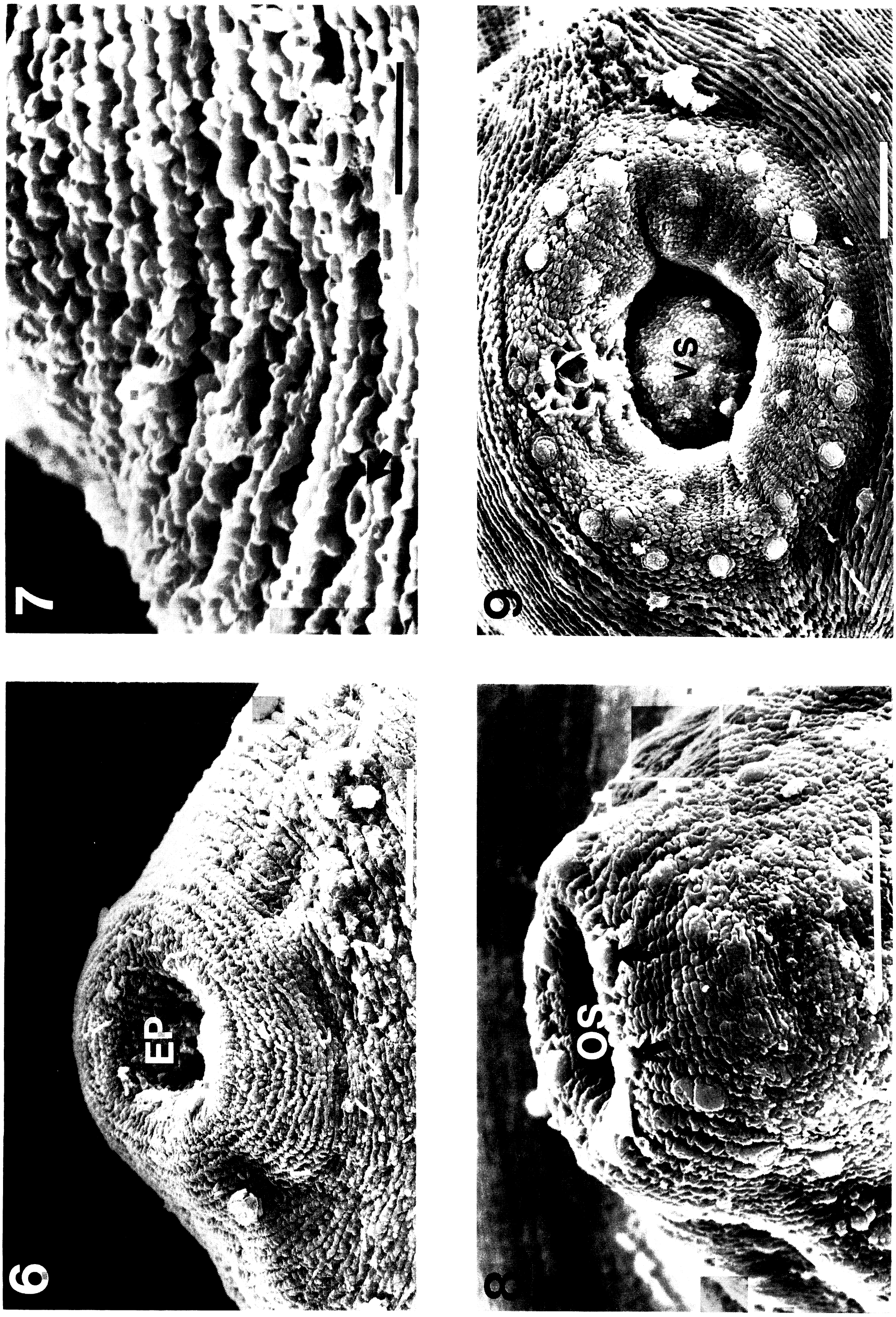
7
9
VS
6
EP
8
OS

Scanning electron microscope studies of surface microtopography of larval Didymozoidae are not available. In this study, it was found that the didymozoid larva has basic morphological and topographical features similar to that of metacercariae of other genetic trematodes that already have been described (Bennett, 1975; Lo *et al.*, 1975; Koie, 1975, 1979, 1985a; Fujino *et al.*, 1979; Hoole and Mitchell, 1981; Fried and Fujino, 1984). However, there are differences in the microtopography of the surface and types and distribution of papillae. Increase in the tegumentary surface in the form of concentrically arranged ridges bearing protuberances, as found on the didymozoid larva, has previously been described for metacercariae of trematodes. Such structural arrangements have been associated with nutrient absorption and/or respiratory gaseous exchange. It is also possible that the ridges may provide the necessary flexibility and expansion needed for locomotion through tissues of the host. Lack of spines on the body surface of the larval didymozoid, as shown in this study, indicates a major topographical transformation compared to the spiny miracidia of some species (Baylis, 1938).

Different types of ciliated and non-ciliated structures with presumed sensory functions have been described on the surfaces of trematode metacercariae, while in this study only a non-ciliated domed papillae was observed. This type is the most prevalent on the oral and ventral suckers of metacercariae that have been examined (Koie, 1985b). It is reasonable to assume that the domed papillae located on the forebody of the larva act as mechanoreceptors functioning during migration through tissues or body cavity of the paratenic and final hosts searching for the definitive site of development. Studies on the number and arrangement of papillae around suckers of adult and larval Didymozoidae may prove to be helpful in solving taxonomical problems in this group of trematodes.

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