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MORPHOLOGICAL OBSERVATIONS ON THE EGG AND FIRST INSTAR LARVA OF *METACUTEREBRA APICALIS* (DIPTERA: CUTEREBRIDAE)

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Descriptions are given of the egg and first instar larva of Metacuterebra apicalis *(Diptera: Cuterebridae) when viewed by light and scanning electronic microscopes.*

Key words: Diptera – Cuterebridae – *Metacuterebra apicalis* – optical microscope – scanning electron microscope – egg and first instar larva – morphological descriptions

Metacuterebra apicalis is a Cuterebrid (Diptera: Cyclorrapha) of the Neotropical Region. Its larvae are parasitic in rodents (Guimarães et al., 1983), causing cutaneous furuncular lesions (myiasis). The life cycle of *M. apicalis*, in its natural host *(Oryzomys subflavus)* and laboratory white rats (albino *Rattus norvegicus*) was recently described (Leite & Williams, 1988).

The eggs and first instar larvae of Cuterebrids, as seen under a light microscope, have been described by several authors (Hadwen, 1915; Townsend, 1915; Dalmat, 1943; Molliett, 1950; Ryckman & Lindt, 1954; Bennett, 1955; Uberlaker & Keller, 1964; Capelle, 1970; Graham & Capelle, 1970; Baird & Graham, 1973). Those of a few species have also been studied by scanning electronic microscope (Baker, 1986; Colwell & Kokko, 1986; Leite, 1988). This paper records morphological observations on the egg and first instar larva of *M. apicalis* by means of both light and scanning electron microscopes.

MATERIALS AND METHODS

The eggs and first instar larvae of *M. apicalis* were obtained in the laboratory when the life cycle of the fly was being studied. Specimens were first examined alive and later killed and fixed in Dietrich/San Jean fluid for 24 hours and stored in 70% ethyl alcohol. Material was processed for scanning electron microscope studies in the manner described by Leite & Lopes (1987).

RESULTS

Twenty-five eggs and 25 larvae were measured after fixation.

Egg. The egg (Figs 1 and 2) is boat-shaped, light brown in colour, 1.10-1.19 ($\bar{x}$ = 1.17 ± 0.27) mm long and 0.17-0.21 ($\bar{x}$ = 0.19 ± 0.01) mm wide. The anterior end is flattened at the micropylar plate (Fig. 3). The dorsal and lateral surfaces are curved; the ventral surface is flattened, and the egg tapers posteriorly. The exo-chorion is sculptured with a network appearing as pits and ridges, high-relief ridges forming irregular four to six sided polygons, sexagons mainly occurring on the dorsal surface. An ellipsoid operculum is situated in an anterio-dorsal position. The operculum bears high-relief ridges except in a posterior pitted area. The micropylar plates (Fig. 4) have a central aperture with prominent walls and surrounded by a few pits. The micropylar aperture is connected to a micropylar canal (Fig. 5) that penetrates the outer layer of the vitelline membrane. The ventral surface of the egg is covered by a substance that, presumably, acts as an adhesive with substrates.

First instar larva. The larva (Figs 9 and 13) is spindle-shaped, broadest at the fifth segment and tapering towards the 12th segment, which has a dorsal expansion. Pale white in colour, it is 0.57-1.18 ($\bar{x}$ = 1.00 ± 0.16) mm long,

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0.13-0.18 ($\bar{x}$ = 0.16 ± 0.01) mm wide. The pseudocephalon (Figs 11, 14 and 20-22) has paired cone-shaped antennae projecting laterally, each bearing a cerdiform papilla. The palps have two papilliform tubercles, and there are hooks (maxillae) on either side of the mouth. Segments 2-10 have backwardly directed spines along the anterior margin. Segment 2 (Figs 14 and 20-22) has small, sharp spines with recurved tips. Trachea extend to the anterior part of the body but anterior spiracles were not seen. The 3rd and 4th segments (Fig. 15) have three types of spines: *a* – long, triangular, transversely implanted, and with sharply pointed tips; *b* – large, triangular, longitudinally implanted, with curved apices; *c* – small, slender, smoothly recurved, and with sharp tips. Sparse, very finely pointed backwardly directed spines are present on half of segment 4. The ventral surface of the three thoracic segments bears a pair of trilobulate sensillae. Segment 5 bears spines like those on the 4th segment. Segments 7-10 (Fig. 16), have small, sharp spines and sparse, finely pointed spines (Fig. 17). The 11th segment has anteriorly directed small spines, of both types, on the dorsal surface (Fig. 18). The 12th segment (Fig. 10) has a dorsal expansion arising near the anus and two dorsally situated posterior spiracles (Figs 19 and 23), each having two openings and a delicate peritreme.

The cephalonpharyngeal skeleton (Figs 24-26) is dark brown in colour, extends from the mouth to the 3rd segment, and is 169 μm long. The maxillae have large bases, bear smooth and pointed ventrally recurved hooks and have a sharp dorsal process. Each maxilla has a very small foramen. The dentate sclerite lies near the base of the maxilla, bears an anterior sharp process, and the two are united by two parallel bars (the tentopharyngeal sclerite). The tentopharyngeal sclerite has a pointed process. There is no clypeal arch. The dorsal cornua are strongly arched and the ventral cornua are straighter.

DISCUSSION

Under the light microscope, the egg of *M. apicalis* is similar to that of *M. baui* (Fonseca, 1940) and to those of eight species of *Cuterebra* (Hadwen, 1915; Townsend, 1915; Dalmat, 1943; Moilliet, 1950; Baird & Graham, 1973; Ryckman & Lindt, 1954; Haas & Dicke, 1958; Penner, 1958; Capelle, 1970; Graham & Capelle, 1970). In some cases, there are marked differences in the colour of the eggs of different species. Thus, Townsend (1915) recorded that the eggs of *C. cuniculi* are salmon pink; those of *M. baui* are orange (Fonseca, 1940); Moilliet (1950) and Baird & Graham (1973) described those of *C. tenebrosa* as creamy-white; the eggs of *D. hominis* are white (Moya Borja, 1966).

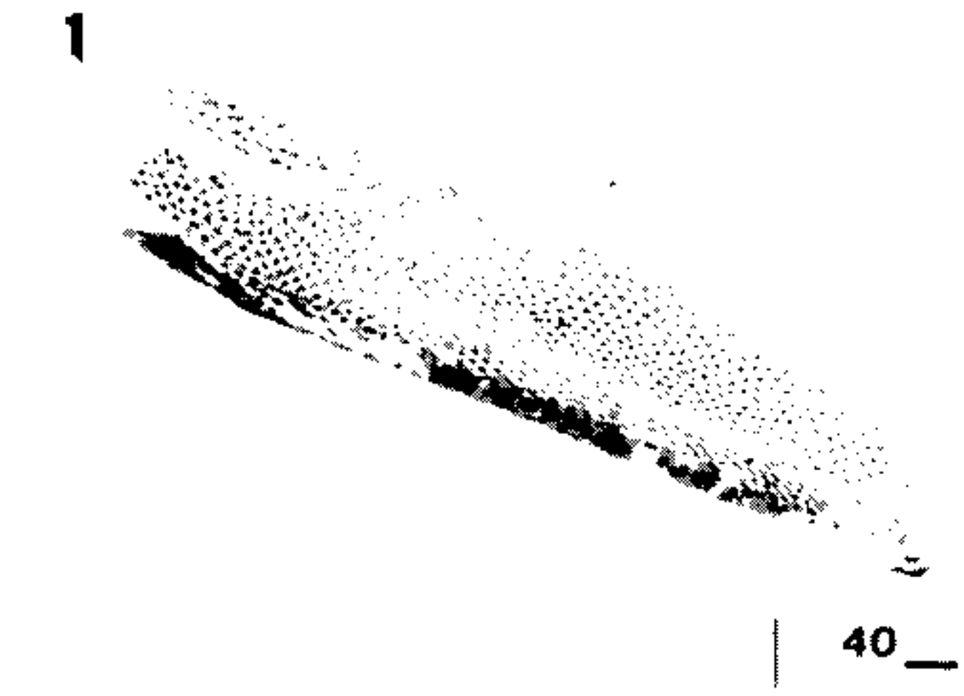
Some morphological differences between eggs can by seen by optical microscopy. The egg of *D. hominis* (1,5 mm long x 0.6 mm wide) is much longer and broader than that of *M. apicalis*. Bennett (1955) and Ubelaker & Keller (1964) recorded the presence of spines on the outer surface of the operculum of *C. emasculator*. Such spines were not detected on the operculum of the eggs of *M. apicalis*.

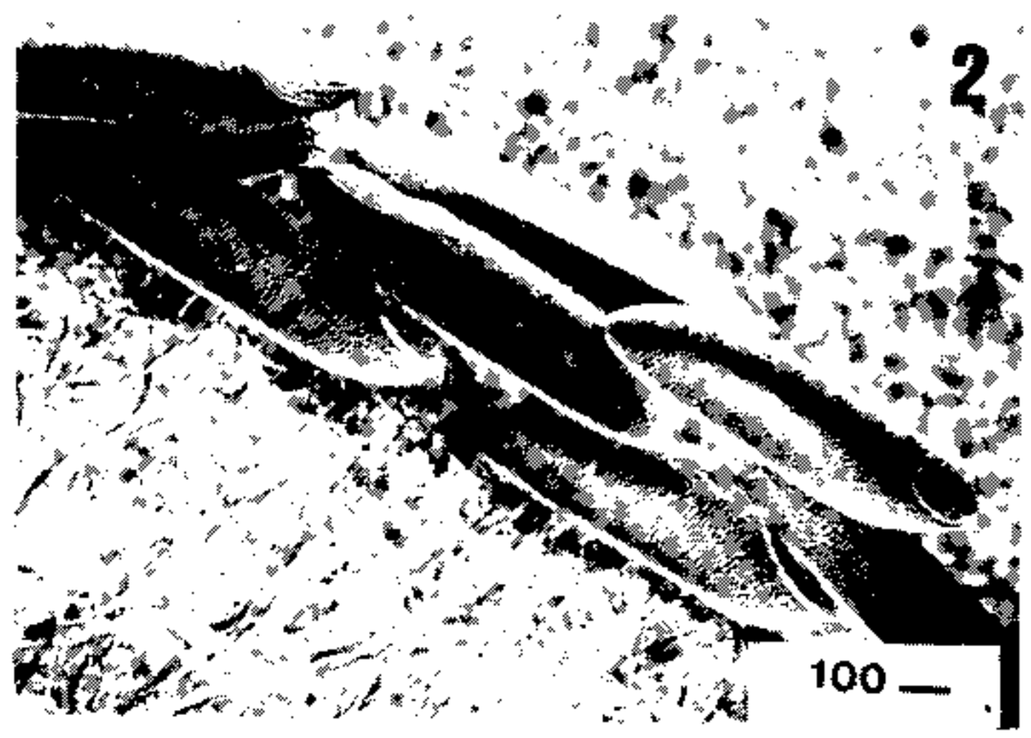
Studying the eggs of *C. horripilum* by scanning electronic microscopy, Baker (1986) described bands of rectangular pits on the exochorion. *M. apicalis* lacks such pits. In comparison with the eggs of *D. hominis* (Leite, 1988), the exochorion of *M. apicalis* is not sculptured with short, high-relief ridges, has a more flattened micropylar plate and lacks larger pits around the micropylar aperture.

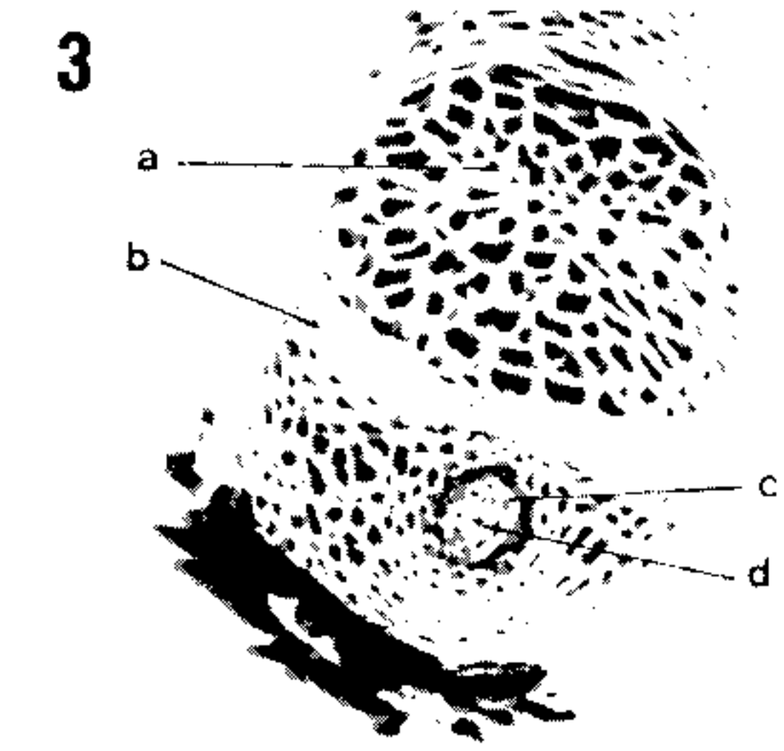
Under the optical microscope, the first instar larva of *M. apicalis* is similar to those of four species of *Cuterebra* (Ryckman & Lindt, 1954; Haas & Dicke, 1958; Penner, 1958; Capelle, 1970) but is much shorter than two other species of *Cuterebra* (Parker & Wells, 1919; Ferris, 1920; Baird & Graham, 1973). The first stage larva of *D. hominis* has large, pointed spines of abdominal segments 5-7, lacks spines on the 8th and 9th segments and has medium pointed, forwarded directed spines on segments 11 and 12 (Surcouf, 1913; Newstead & Potts, 1925; Moya Borja, 1966). The first stage larva of *M. apicalis* differs in all three characters.

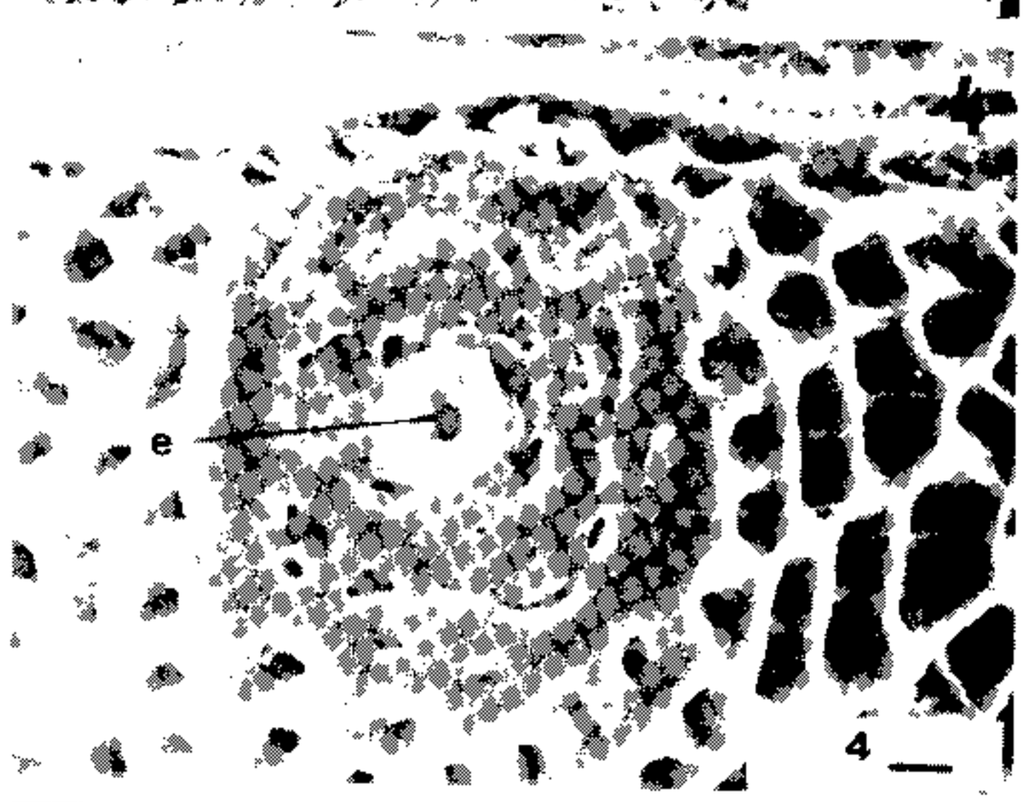
The cephalonpharyngeal skeleton of *M. apicalis* is shorter than that of *C. emasculator* (Bennett, 1955) and *D. hominis* (Moya Borja, 1966), and the tentopharyngeal sclerite of *M. apicalis* is smaller than that of *C. tenebrosa* (Baird & Graham, 1973). Like the first stage larva of *D. hominis* (Lopes, 1982), that of *M. apicalis* lacks mandible and clypeal arches.

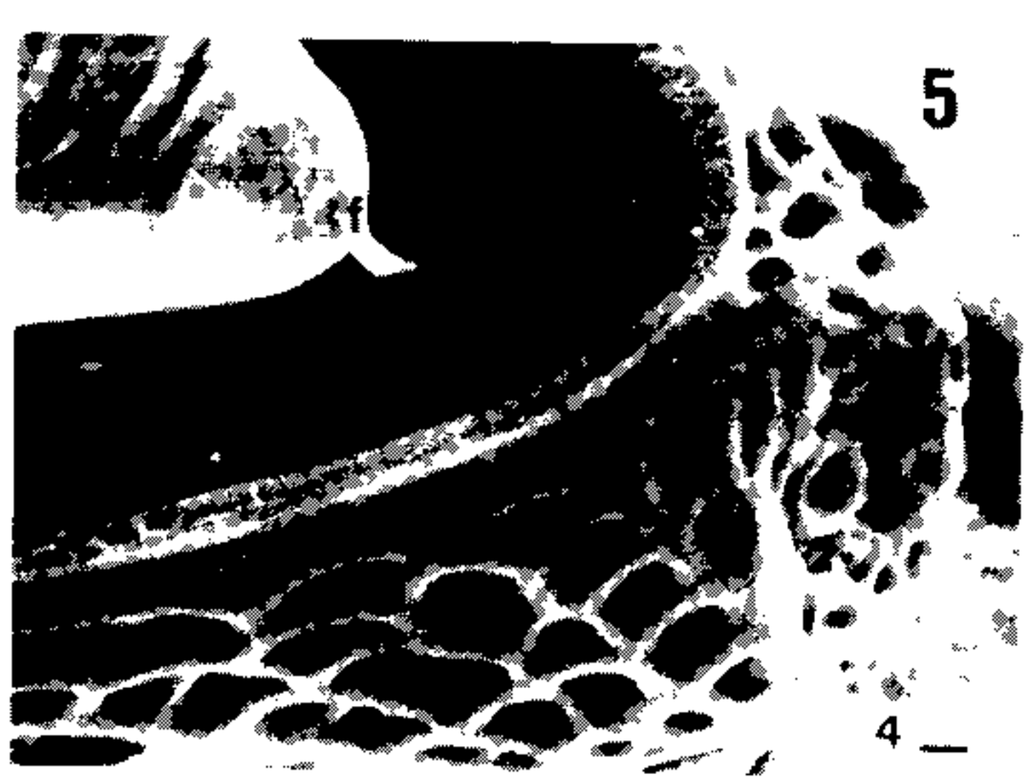
By scanning electronic microscopy, the head of the first instar larva of *M. apicalis*, apart from many well-defined ridges on the maxillae

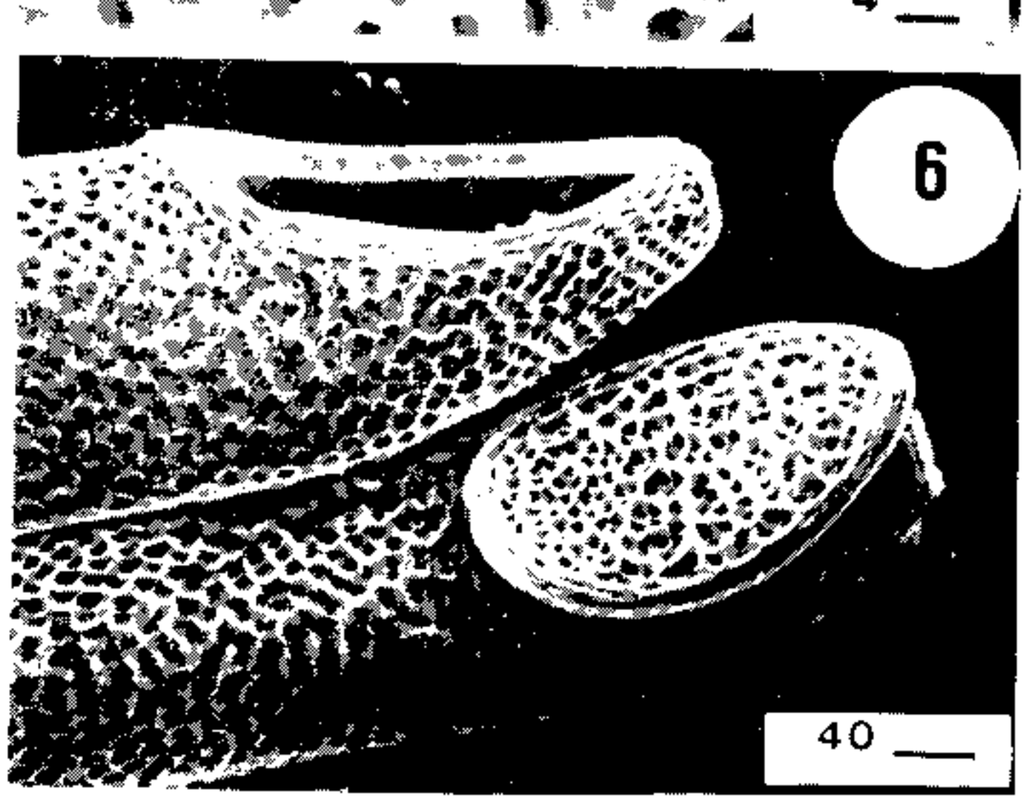


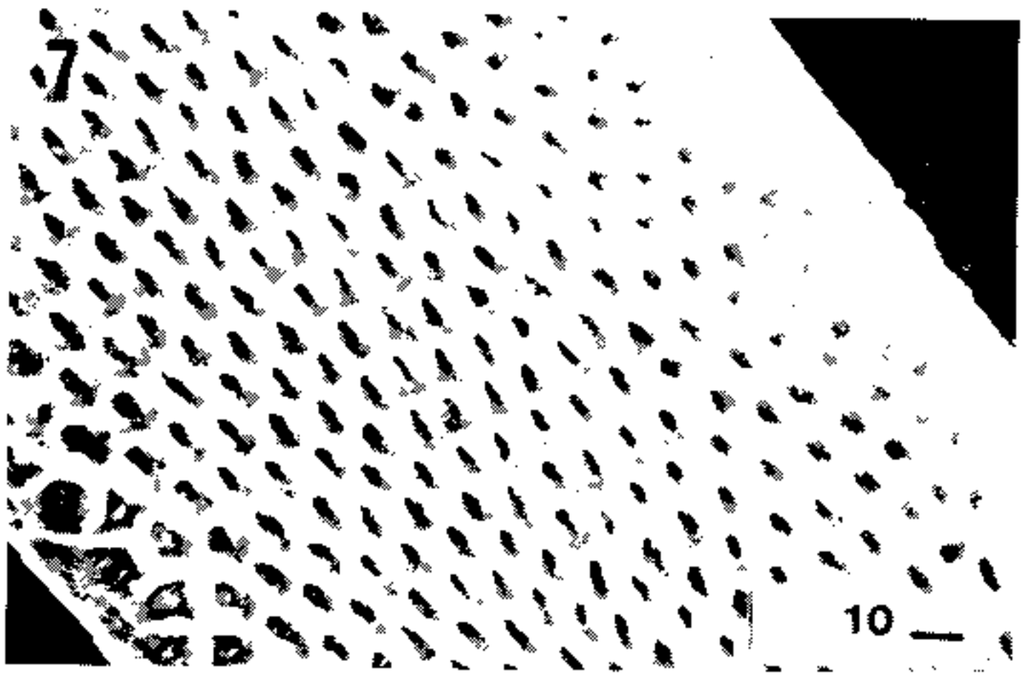


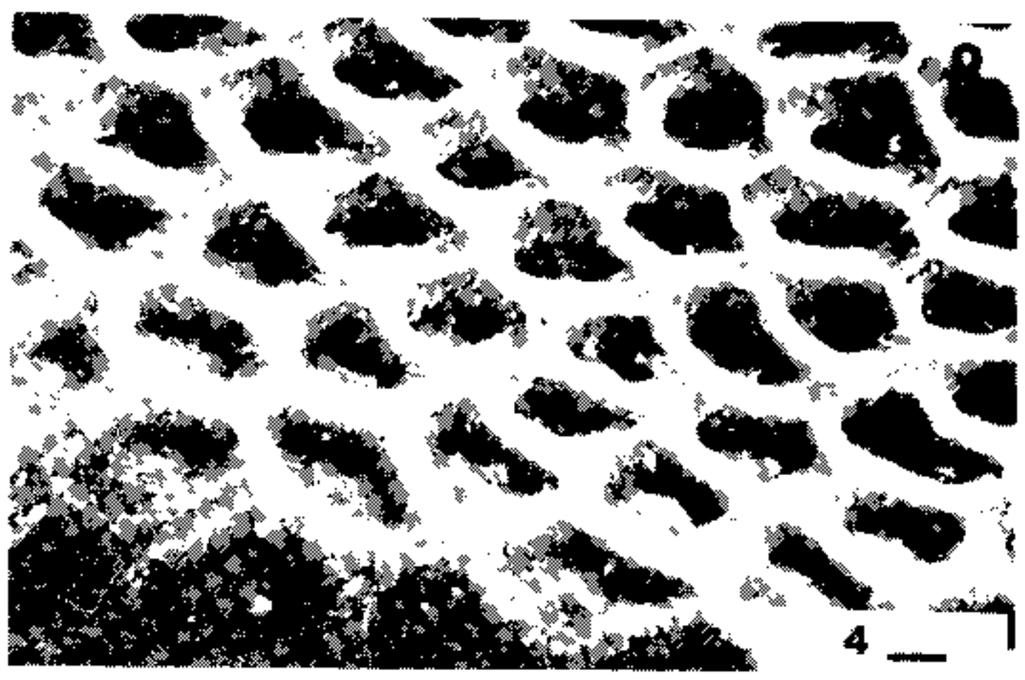




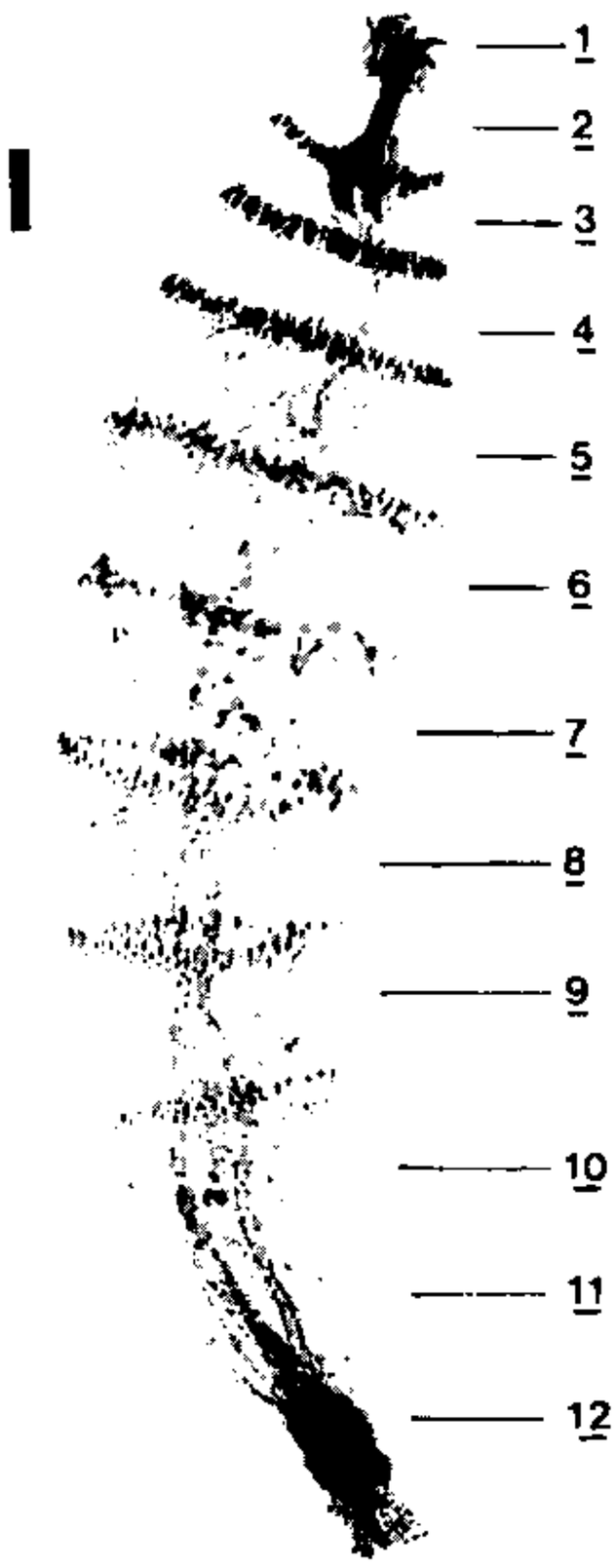




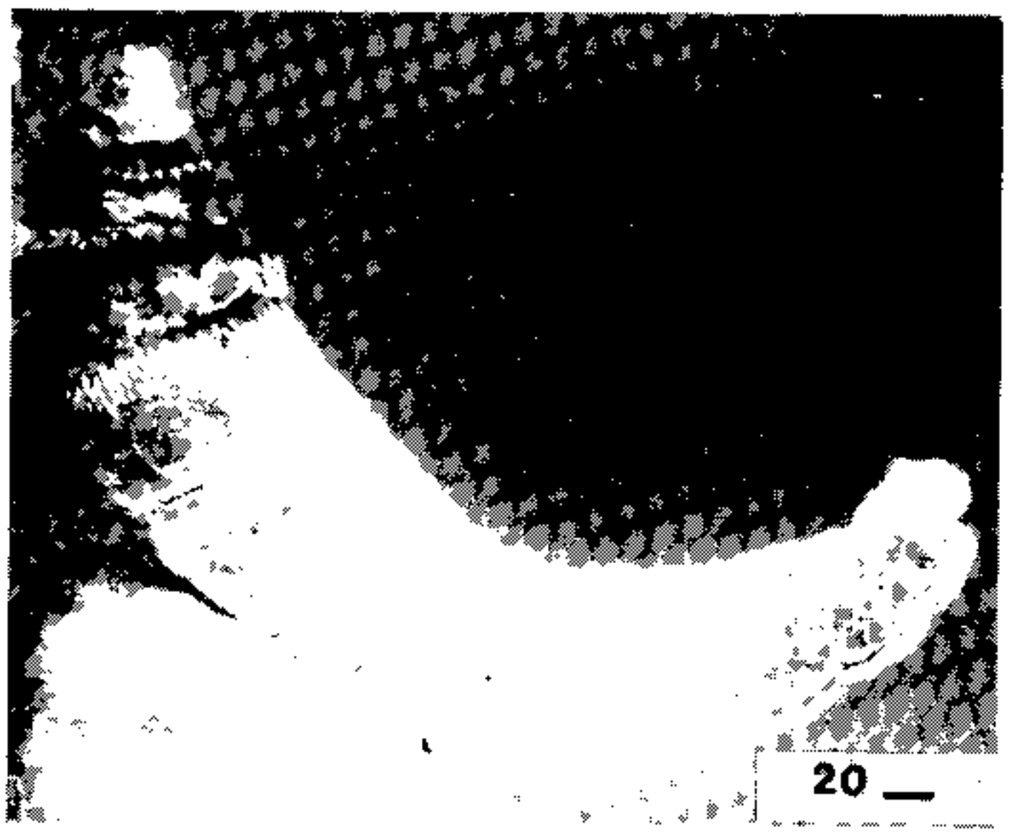


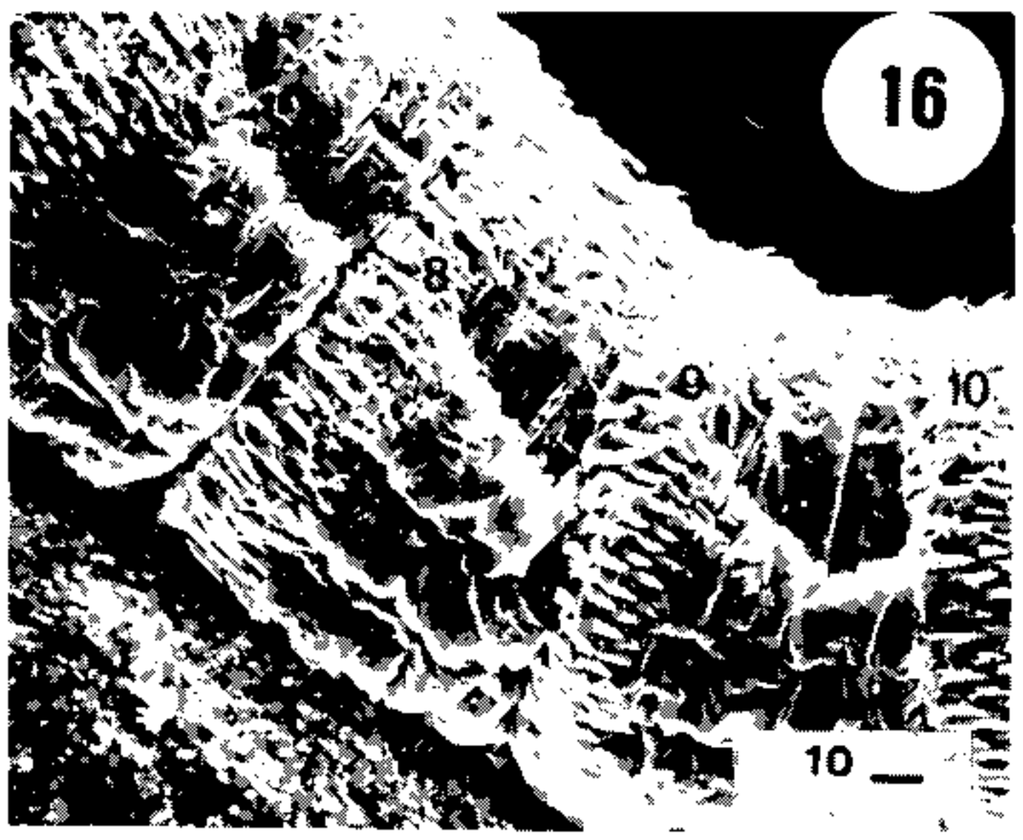


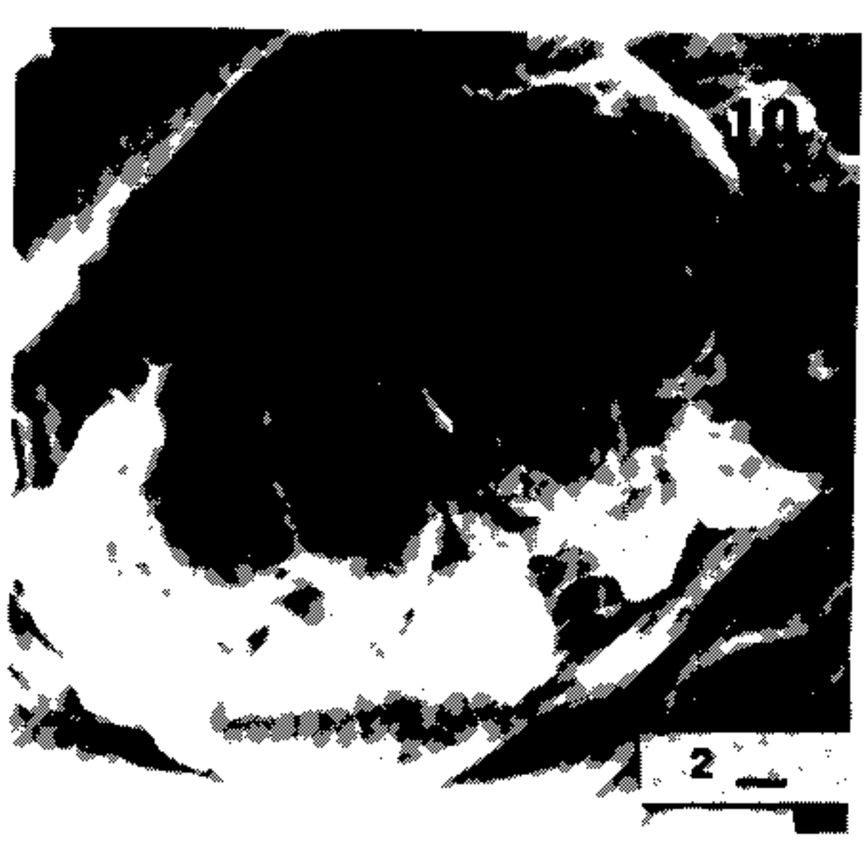
Scanning electron microscope photomicrographs of the egg of *Metacuterebra apicalis*. Fig. 1: dorsal view. Fig. 2: overlapping row of eggs. Fig. 3: view of anterior end (showing: a – operculum; b – hatching line of the operculum; c – micropylar plate; d – micropyle). Fig. 4: micropylar plate and micropyle (e). Fig. 5: open operculum, showing (f) micropylar canal passing through the vitelline membrane. Fig. 6: anterior end of shell before (below) and after (above) eclosion. Fig. 7: apertures of the aerophyle. Fig. 8: ventro-lateral view, showing exo-chorion covered with a adhesive substance. Scales lines in μm.



Light microscope photomicrographs of the first instar larve of *Metacuterebra apicalis*. Fig. 9: complete larva in dorsal view, with segments numbered sequentially. Fig. 10: lateral view of posterior segments (scale line = 0.1 nm), indicating trachea (g), posterior spiracles (h) and dorsal expansion (i). Fig. 11: lateral view of anterior end, indicating antennal (j) and cephalopharyngeal skeletion (k). Fig. 12: posterior tip in dorsal view. Scale line in 0.1 mm.







Scanning electron microscope photographs of the first instar larva of *Metacuterebra apicalis*. Fig. 13: complete larva in lateral view. Fig. 14: frontal view of anterior end. Fig. 15: thoracic and abdominal segments in dorsal view. Fig. 16: lateral view of segments 7-10. Fig. 17: delicate spine between 4th and 5th segments. Fig. 18: arrangement of spines on the dorsal surface of the 12th segment. Fig. 19: peritreme of the posterior spiracle. Scale lines in μn.

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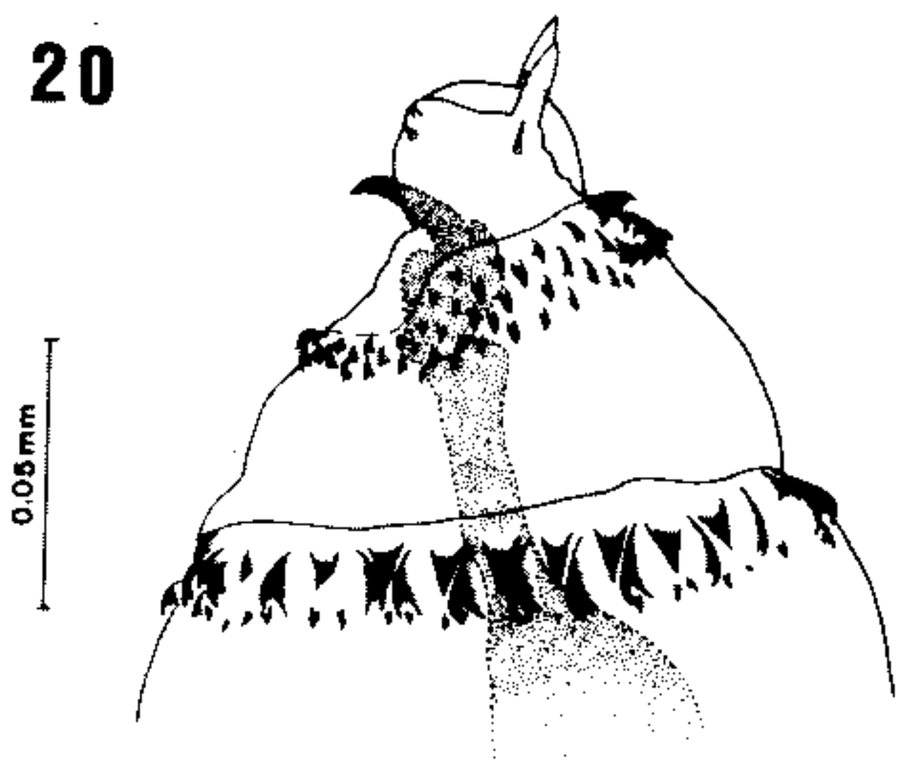

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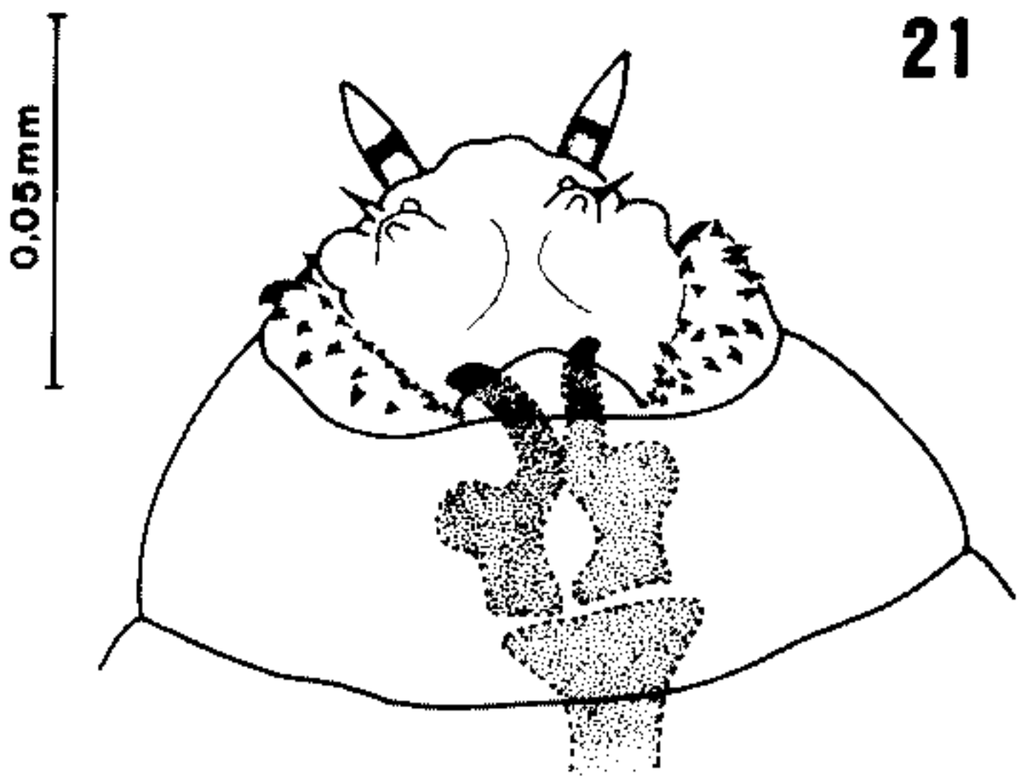

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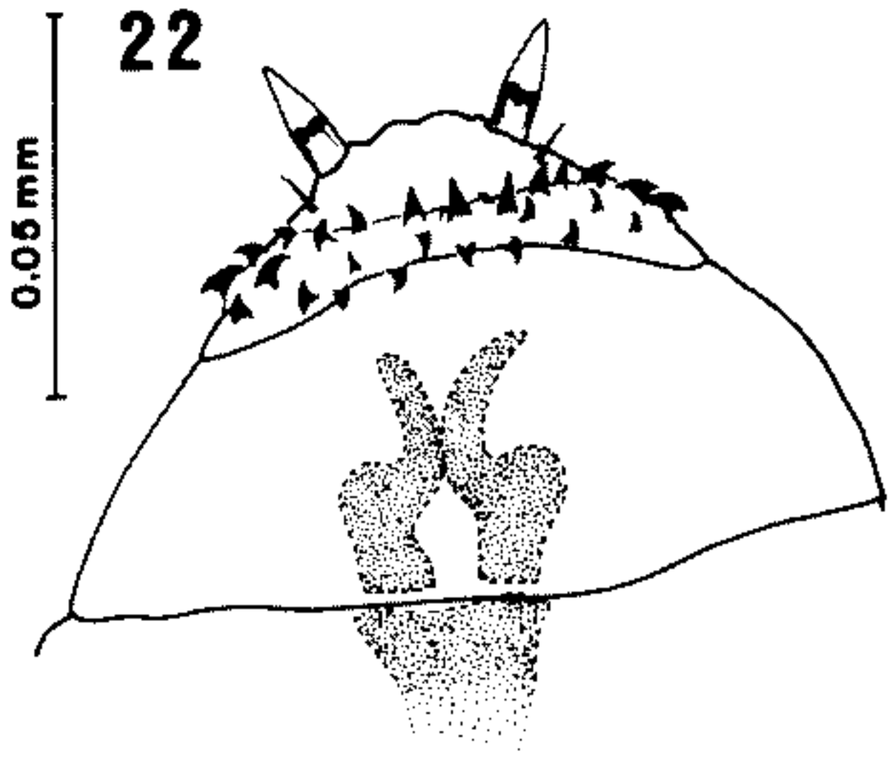

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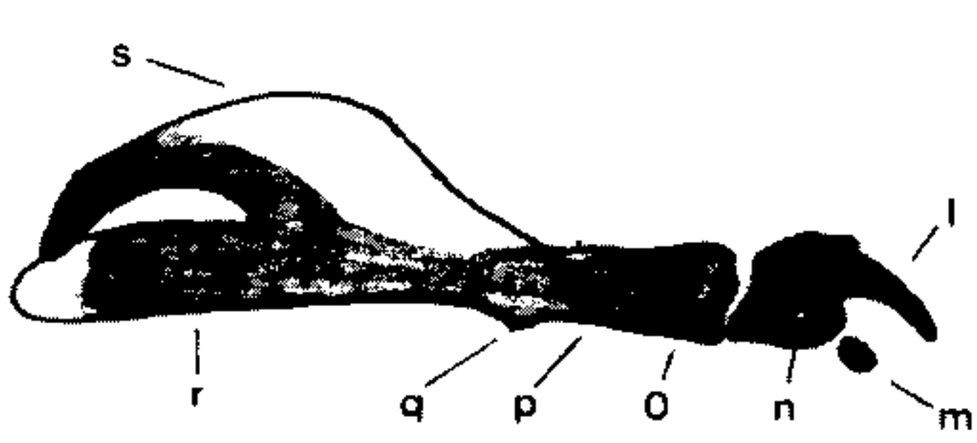

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Camera lucida drawings of the first instar larva of *Metacuterebra apicalis* viewed under a light microscope. Fig. 20: lateral view of anterior end. Fig. 21: ventral view of anterior end. Fig. 22: dorsal view of anterior end. Fig. 23: posterior spiracles. Fig. 24: cephalonpharyngeal skeleton in lateral view (indicating: *l* – maxilla; *m* – dentate sclerite; *n* – foramen; *o* – hypopharyngeal sclerite; *p* – tentopharyngeal sclerite; *g* – lateral process; *r* – ventral cornu; *s* – dorsal cornu). Fig. 25: cephalopharyngeal skeleton in ventral view. Fig. 26: cephalopharyngeal skeleton in dorsal view.

(Baker, 1986) is similar to that of *C. horripilum.* In general, the body of the first instar larva of *M. apicalis* resembles that of *C. fontinella* (Colwell & Kokko, 1986). The differences between the first stage larvae of *M. apicalis* and *D. hominis,* already mentioned, are more apparent when they are viewed by scanning electronic microscopy.

The apparent absence of an anterior spiracle in the first stage larva of *M. apicalis* could be an artefact because the first thoracic segment was retracted in all material examined. A small spiracle has been described from certain other Cylcorraphans (Kitching, 1976; Cantrell, 1981; Leite et al., 1985).

RESUMO

Observações morfológicas do ovo e da larva de primeiro estágio de *Metacuterebra apicalis* (Diptera: Cuterebridae) – O ovo e a larva de primeiro estágio de *Metacuterebra apicalis* (Diptera, Cuterebridae) são descritos a nível de microscopia óptica e eletrônica de varredura.

Palavras-chave: Diptera – Cuteribridae – *Metacuterebra apicalis* – microscopia óptica – microscopia electrônica de varredura – ovo – larva de primeiro estágio – descrição morfológica

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REFERENCES

BAIRD, C. R. & GRAHAM, C. L., 1973. *Cuterebra tenebrosa:* description of immature stages and a redescription of the adult (Diptera: Cuterebridae). *Can. Entomol., 105:* 1281-1293.

BAKER, G. T., 1986. Morphological aspects of the egg and cephalic region of the first instar larva of *Cuterebra horripilum* (Diptera: Cuterebridae). *Wasmann J. Biol., 44:* 66-72.

BENNETT, G. F., 1955. Studies on *Cuterebra emasculator* Fitch 1856 (Diptera: Cuterebridae) and a discussion of the status of the genus *Cephenemyia* Ltr. 1818. *Can. J. Zool., 33:* 75-98.

CANTRELL, B. K., 1981. The immature stages of some Australian Sarcophagidae (Diptera: Sarcophagidae). *J. Aust. Entomol. Soc., 20:* 237-248.

CAPELLE, K. J., 1970. Studies on the life history and development of *Cuterebra polita* (Diptera: Cuterebridae) in four species of rodents. *J. Med. Entomol., 7:* 320-327.

COLWELL, D. D. & KOKKO, E. G., 1986. Preparation of larvae for scanning electron microscopy using a freeze-substitution technique. *Can. J. Zool., 64:* 797-799.

DALMAT, H. T., 1943. A contribution to the knowledge of the rodent warble flies (Cuterebridae). *J. Parasitol., 29:* 311-318.

FERRIS, G. F., 1920. The first stage larvae of *Cuterebra americana* (Fabr.) (Diptera: Oestridae). *Psyche, 27:* 13-14.

FONSECA, F., 1940. Nova espécie de *Oricuterebra* do Brasil. (Diptera: Oestridae). *Rev. Entomol., 11:* 662-671.

GRAHAM, C. L. & CAPELLE, K. J., 1970. Redescription of *Cuterebra polita* (Diptera: Cuterebridae) with notes on its taxonomy and biology. *Ann. Entomol. Soc. Am., 63:* 1569-1573.

GUIMARÃES, J. H.; PAPAVERO, N. & PRADO, P. P., 1983. Família Cuterebridae (exceto *Dermatobia hominis*). In: As miíases na Região Neotropical (identificação, biologia e bibliografia). *Rev. Bras. Zool., 1:* 354-414.

HAAS, G. E. & DICKE, R. J., 1958. On *Cuterebra horripilum* Clark (Diptera: Cuterebridae) parasitizing cottontail rabbits in Wisconsin. *J. Parasitol., 44:* 527-540.

HADWEN, S., 1915. A description of the egg and ovipositor of *Cuterebra fontinella,* Clark. (Cottontail Bot.). *Proc. Entomol. Br. Columbia, 5:* 88-91.

KITCHING, R. L., 1976. The immature stages of the Old-World screw-worm fly, *Chrysomya bezziana* Villeneuve, with comparative notes on the Australasian species of *Chrysomya* (Diptera: Calliphoridae). *Bull. Entomol. Res., 66:* 195-203.

LEITE, A. C. R., 1988. Scanning electron microscopy of the egg and the first instar larva of the *Dermatosia hominis* (Diptera: Cuterebridae). *Mem. Inst. Oswaldo Cruz, 83:* 253-257.

LEITE, A. C. R.; GUEVARA, J. D.; GUIMARÃES, M. P. & LIMA, W. S., 1985. Ultra-estrutura da larva de primeiro estágio de *Cochliomyia hominivorax* (Coquerel, 1858) (Diptera: Calliphoridae). In: Congresso da Sociedade Brasileira de Parasitologia, 9, Fortaleza. *Resumos,* p. 145.

LEITE, A. C. R. & LOPES, H. S., 1987. Second contribution to the knowledge of the larvae of the Raviniini (Diptera: Sarcophagidae) based on observations using scanning electron microscope. *Mem. Inst. Oswaldo Cruz, 82:* 219-226.

LEITE, A. C. R. & WILLIAMS, P., 1988. Life cycle of *Metacuterebra apicalis* (Diptera: Cuterebridae). *Mem. Inst. Oswaldo Cruz, 83:* 485-491.

LOPES, H. S., 1982. The importance of the mandible and clypeal arch of the first instar larvae in the classification of the Sarcophagidae (Diptera). *Rev. Bras. Entomol., 26:* 293-326.

MOILLIET, T. K., 1950. Some preliminary observations on the life history of *Cuterebra tenebrosa* Coquillet. *Proc. Entomol. Soc. Br. Columbia, 46:* 1-3.

MOYA BORJA, G. E., 1966. *Estudios sobre la biología, morfología y esterilización del torsalo, Dermatobia hominis* (L., Jr.). Tesis, Magister Scientiae. Turrialba, Instituto Interamericano de Ciencias Agrícolas de la O. E. A. Centro de Enseñanza e Investigación, 63 p.

NEWSTEAD, R. & POTTS, W. H., 1925. Some charac-

teristics of the first stage larvae of *Dermatobia hominis* Gmelin. *Ann. Trop. Med. Parasitol., 19:* 247-260.

PARKER, R. R. & WELLS, R. W., 1919. Observations on and experiments with *Cuterebra tenebrosa* Coquillet. *J. Parasitol., 5:* 100-104, pl. 8.

PENNER, L. R., 1958. Concerning a rabbit cuterebrid, the larvae of which may penetrate the human skin (Diptera: Cuterebridae). *J. Kans. Entomol. Soc., 31:* 67-71.

RYCKMAN, R. E. & LINDT, C. C., 1954. *Cuterebra lepivora* reared from *Sylvilagus audubonii sancti-diegi* in San Bernardino County, California. *J. Econ. Entomol., 47:* 1146-1149.

SURCOUF, M. J., 1913. La transmission du ver macaque par un moustique. *C. R. Hebd. Séanc. Acad. Sci. Paris, 156:* 1406-1408.

TOWNSEND, C. H. T., 1915. On the reproductive and host habits of *Cuterebra* and *Dermatobia. Science, 42:* 253-255.

UBELAKER, J. E. & KELLER, B. L., 1964. Observations of eggs and larvae of *Cuterebra emasculator* (Fitch, 1856). *Trans. Kans. Acad. Sci., 67:* 713-715.