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# MORPHOLOGICAL AND HISTOCHEMICAL DEMONSTRATION OF HOOKS IN THE ROSTELLAR REGION OF CYSTICERCUS BOVIS

Z. ŽĎÁRSKÁ

Institute of Parasitology, Czechoslovak Academy of Sciences, Prague

**Abstract.** Hooklets and early developmental stages of hooks were discovered in the tegument among the microtriches in the rostellar region of 83- and 108-day-old *Cysticercus bovis*. The cortical layer of hooks consists of proteins with tyrosine and SS groups. The rostellar sac and bulb are distinctly developed in the parenchyma of the rostellar region.

The present paper follows the previous ones dealing with the tegument and parenchyma of *C. bovis* (Žďárská 1973, 1975). Although it is known that the adult cestode *Taeniarhynchus saginatus* has no hooks, we have determined the hooks in its larva, *C. bovis*. The presence of these hooks in the rostellar region of *C. bovis* has not been noticed by any author.

In our studies we used the same histological and histochemical methods as in the previous papers (Žďárská 1973, 1975). The material originated from experimental infections of calves performed by Dr. Machnicka from the Polish Academy of Sciences, Warszaw (Machnicka et al. 1976).

## RESULTS

The rostellar region of 83- and 108-day-old *C. bovis* is situated at the end of the spiral canal in the centre between four suckers. If the scolex is inverted, then it lies above the suckers, but if the scolex is everted, the rostellar region is localized between the suckers at the level of their distal end.

The shape of the rostellar region depends on the form of the scolex (inverted or everted). This region is wider than long (Plate I, Figs. 1 and 4; Plate III, Fig. 1) if the suckers of the scolex are inverted and extended along the longitudinal axis (Plate I, Fig. 2; Plate II, Figs. 1, 3, 4) if the suckers are everted.

Two portions of the rostellar region may be distinguished, namely tegument and parenchyma (Fig. 1). The tegument of the rostellar region compared with the tegument of spiral canal is thinner and measures 3μ. Among the microtriches there are isolated hooklets measuring 2.5—4 μ in length and 0.5 μ in width (Plate III, Figs. 1, 2, 3) which stain totally with Mallory's phosphotungstic haematoxylin. The apical part of rostellum is well visible (Plate II, Figs. 2, 4; Plate IV, Fig. 2). It contains minute granules (staining intensely with Mallory's phosphotungstic haematoxylin — Plate IV, Figs. 1, 2) and hooks 6.5—7.5 μ long and 1.5—2 μ wide (Plate IV, Figs. 1, 2, 3, 4) which, unlike the hooklets, are formed by two layers, cortex and medulla. The cortical layer of these hooks is stained intensely with Mallory's phosphotungstic haematoxylin, while the medullar part remains unstained.

In the parenchymal part of the rostellar region the rostellar sac, bulb and praebulb may be distinguished (Fig. 1). The rostellar sac is formed by longitudinal and circular

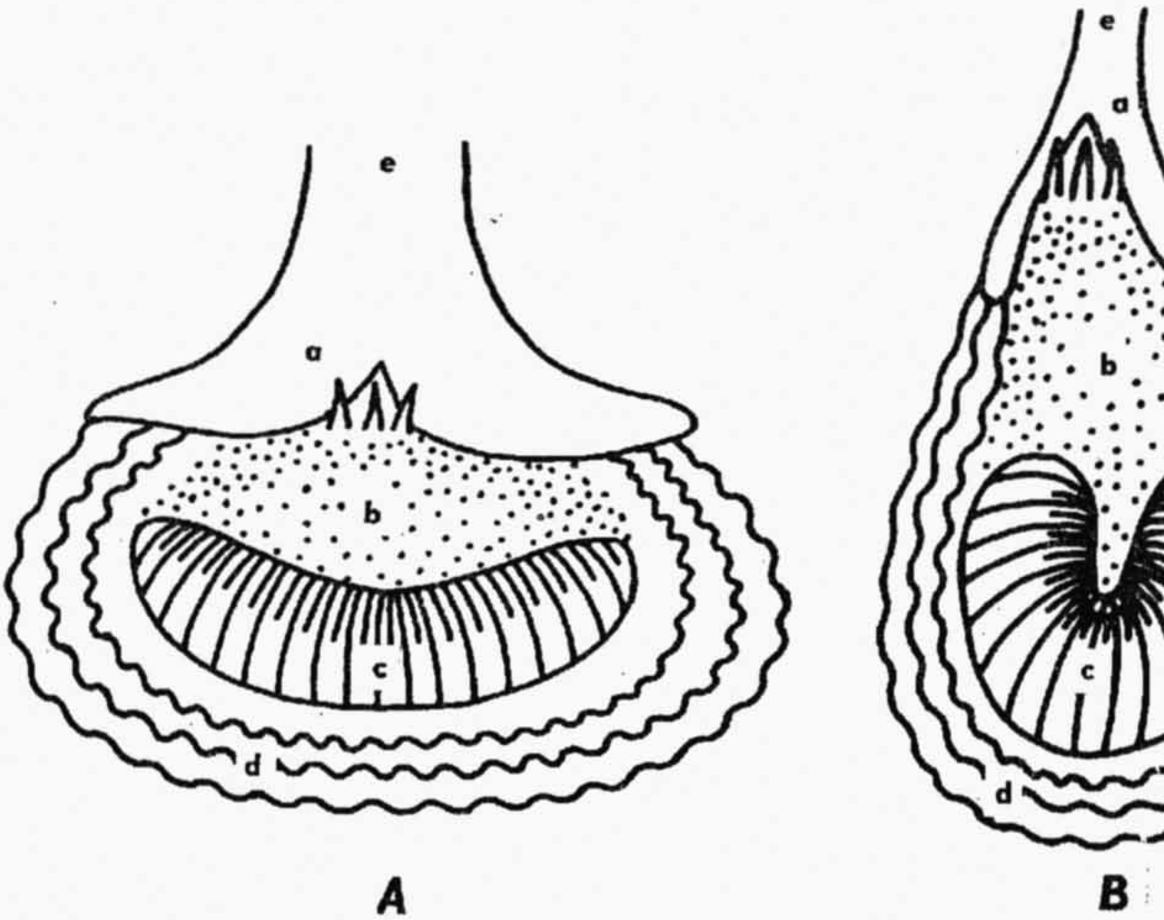


**Fig. 1.** *C. bovis* scheme of rostellum in scolex with inverted (A) and everted (B) suckers. a — apical part and hooks, b — praebulb, c — bulb, d — rostellar sac., e — spiral canal.

fibres (Plate II, Figs. 3, 4). Its shape depends on the form of the scolex (inverted or everted). In our material, a majority of scolices were inverted, but occassional scolices with everted suckers were also found. In scolices with inverted suckers the rostellar sac is dish-shaped (Plate III, Fig. 1), while in scolices with everted suckers, the rostellar sac has the form of an elongate pouch (Plate I, Fig. 2; Plate II, Figs. 3, 4). In everted scolices the wall of the spiral canal is not everted completely; its uppermost portion remains inverted and forms a narrow canal between the four everted suckers, which leads to the rostellum (Plate I, Fig. 2; Plate II, Figs. 1, 3, 4).

The bulb consists of dense muscular fibres. Its shape depends on the pressure inside the scolex. In scolices with inverted suckers (Plate I, Figs. 1, 4; Plate III, Fig. 1) the bulb is flat (100 $\mu$ wide and 30 $\mu$ long), since the layer of the connective tissue (receptaculum) on the surface does not allow it to extend along its longitudinal axis. In the scolex with everted suckers (Plate II, Fig. 1) the bulb is pressed by the suckers and gets a spherical to oval shape (length 80 $\mu$, width 68 $\mu$) (Plate II, Figs. 3, 4). In this case the apical part of the rostellum protruding into the spiral canal is pyramid-shaped (Plate II, Fig. 4). In scolices with inverted suckers the apical part of the rostellum is flat. The prebulb is of minute size.

The granules in the apical part of the rostellum and the cortical layer of hooks gave highly positive reactions in Morel-Sisley's method for the detection of tyrosine (Plate IV, Figs. 3, 4) and PAA + aldehyde fuchsine method for the detection of SS groups (Plate IV, Fig. 2). These results supply evidence that both the hooks and granules in the apical part of rostellum contain scleroprotein. The reactions for neutral and acid mucosubstances and lipids were negative.

## DISCUSSION

Our observations show that the structure of the rostellum and rostellar hooks in early developmental stages of *C. bovis* is similar to that of other cestode larvae (Schaaf 1905, Young 1908, Gläser 1909, Crusz 1947, 1948, Baron 1968, Bilqees and Freeman 1969, Mount 1970, Shield et al. 1973). The development of *C. bovis* hooks, however, stops at a certain stage. Crusz (1948) distinguishes 3 stages in the morphogenesis of hooks: development of the blade, thickening of the blade and development of the handle and guard. Our studies reveal that only the first two stages are realized in the morphogenesis of *C. bovis* hooks and no development of handle and guard occurs. According to Mount (1970) the rostellar hooks arise from the enlarged specialized microtriches. The author assumes that the distal portion of the enlarged microtriche serves as a frame on which the secondary thickening takes place. Subsequently, hook protein is deposited along the edges of theses microtriches to form the blade of the definitive hook. This is consistent with the finding of Gläser (1909) and Šlais (1970) that the definitive hook originates by apposition of the substance on the surface of the primitive hook and not by deposition on the inner surface as Leuckart (1874—1886) assumed. In Mount's (1970) opinion the hooks do not originate either de novo as mentioned by Baron (1968) and Bilqees and Freeman (1960) or from a fusion of microtriches after Young (1908) and Crusz (1947). Since the development of hooks of *C. bovis* stops at the beginning of thickening of the blade, it is understandable why the hook organ is not so well developed as in other species of cysticerci (Bilqees and Freeman 1969, Mount 1970, Šlais 1970). In *C. bovis*, the portion corresponding to the rudimentary hook organ is named by us "apical part of the rostellum". This part in *C. bovis*, similarly as the hook organ in other cestode larvae, contains granules staining intensely in Mallory's phosphotungstic haematoxylin. Besides a large number of microtriches, single hooklets were found in the tegument of the rostellar region of *C. bovis*. They were identical with the hooklets of other cysticerci described by Young (1908), Gläser (1909), Crusz (1947—1948), Bilqees and Freeman (1969) and Mount (1970). The microtriches in the rostellar region have already been observed by Mount (1970) in his electron microscopical studies of the cysticercus of *T. crassiceps*. In the opinion of Baron (1968) and Bilqees and Freeman (1969), however, the microtriches are lacking in this cysticercus.

Since the hooks of *C. bovis* do not continue to develop, also the musculature of the rostellar pad and rostellar sac does not develop to such an extent as in other cysticerci. The tegumental parts of the rostellar region (hooklets, hooks and apical part of rostellum) disappear in older stages of *C. bovis*. The parenchymal part of the rostellum, though in rudimentary form, persists even in the adult cestode (Abuladze 1964).

Small granules found in the apical part of the rostellum in *C. bovis* have the same histochemical properties as the cortical layer of hooks. This supports the hypothesis of Bilqees and Freeman (1969) and Mount (1970) that the substance contained in the "granules" in the hook organ (Bilqees and Freeman) probably equivalent to "ribosomal aggregations" (Mount) is deposited on the surface of the hook-blade and forms thus its cortical layer. It was determined by histochemical methods that this layer is formed by a keratin-like protein (Dollfus 1942, Crusz 1947, Gallagher 1964, Dvorak 1969, Šlais 1970). The cortical layer of the hook of *C. bovis* contains scleroprotein rich in tyrosine and SS groups. In the main, the histochemical properties of *C. bovis* hooks are identical with histochemical properties of the cortical layer of the blade of definitive hooks in other taeniid larvae.

## МОРФОЛОГИЧЕСКОЕ И ГИСТОХИМИЧЕСКОЕ ДОКАЗАТЕЛЬСТВО КРЮЧЬЕВ В ОБЛАСТИ ХОБОТКА *CYSTICERSUS BOVIS*

З. Ждярска

**Резюмс.** Среди микротрихов тегумента в области хоботка *C. bovis* в возрасте 83 и 108 дней были обнаружены крючки и ранние стадии развития крючьев. Наружный слой крючьев состоит из белков с тирозином и дисульфидными группами. В паренхиме области хоботка четко развитый мешок хоботка и бульбус.

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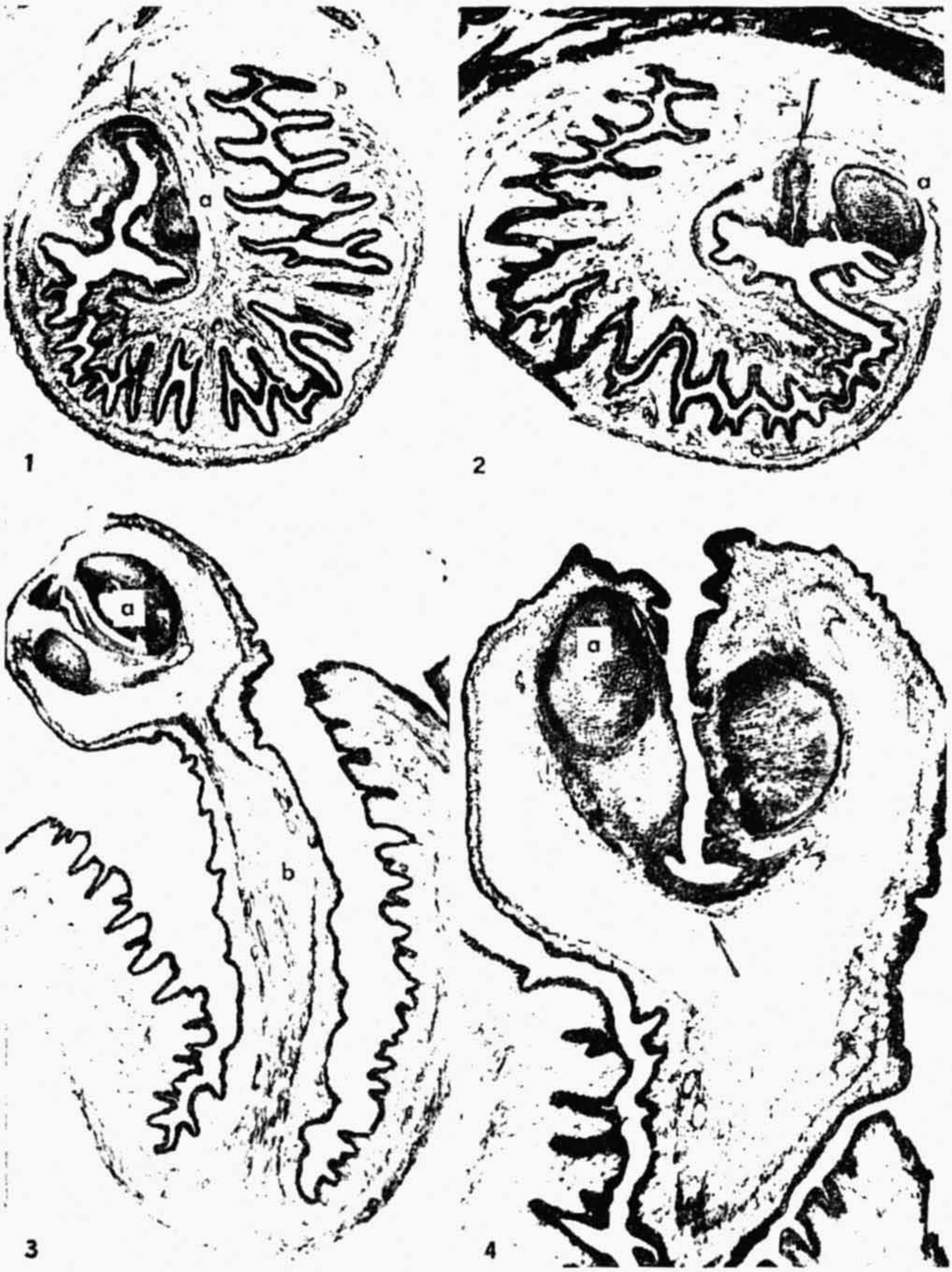
Z. Ž., Parasitologický ústav ČSAV,
Flemingovo n. 2., 166 32 Praha 6,
ČSSR




Location of rostellum in different forms of *Cysticercus bovis* 108 days after infection.
**Arrow** — rostellum, a — sucker, b — neck. Mallory's phosphotungstic haematoxylin.
**Fig. 1.** *C. bovis* with inverted scolex from the musculature of shoulder. (50 ×).
**Fig. 2.** *C. bovis* with partly everted suckers. Musculature of foreleg. (50 ×).
**Fig. 3.** *C. bovis* with outgrown neck and inverted scolex. Musculature of masseter. (50 ×).
**Fig. 4.** The same as in Fig. 3 at higher magnification. (90 ×).

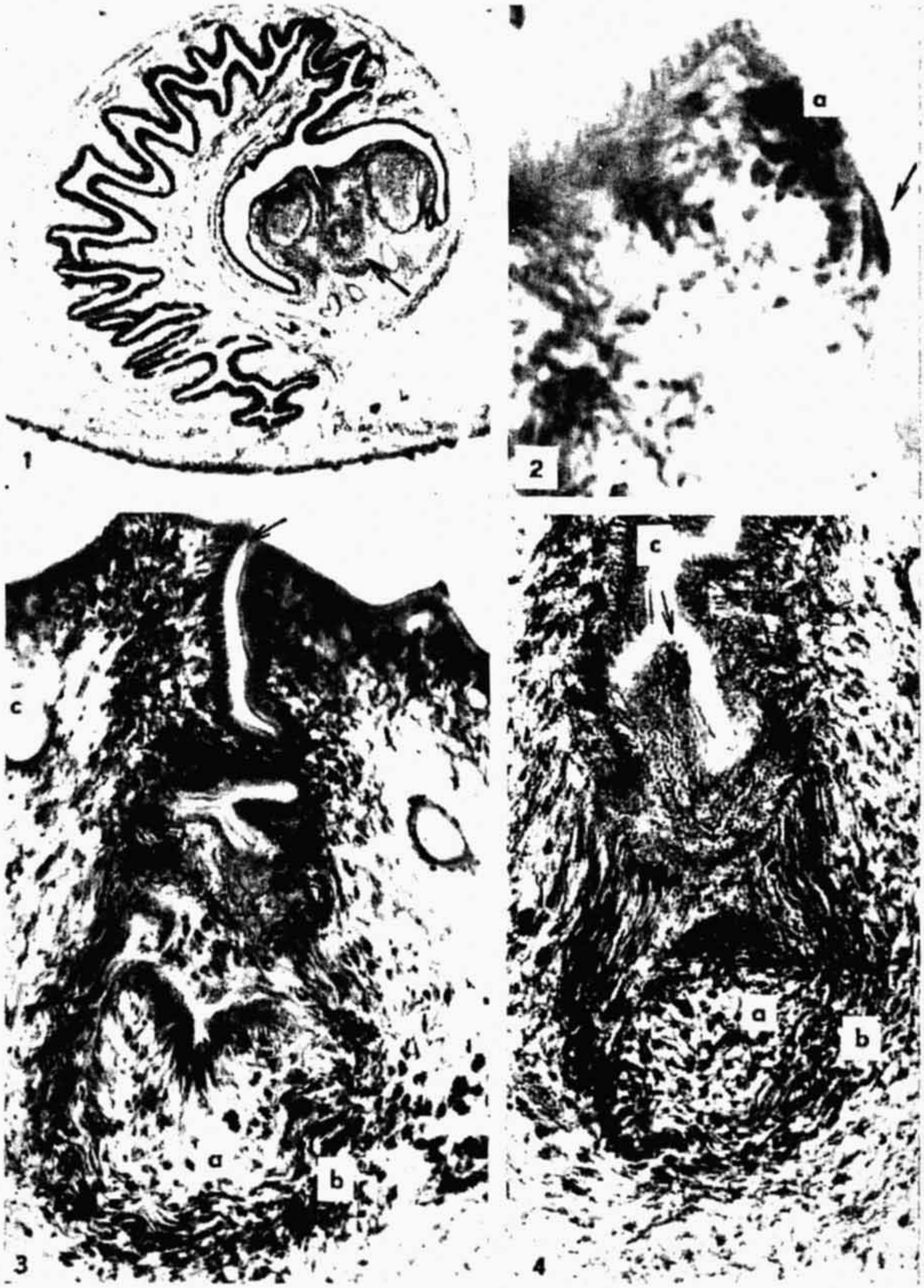



**Fig. 1.** *C. bovis* from musculature of foreleg with fully everted suckers and well visible **rostellar** region (arrow). Masson's blue trichrome. (50×).
**Fig. 2.** Detail of the apical part of rostellum from Fig. 4 with well visible hook (arrow) and **granules** (a). Mallory's phosphotungstic haematoxylin. (2000×).
**Fig. 3.** Longitudinal section through the rostellar region of *C. bovis* with partly everted suckers. Musculature of foreleg. Arrow — spiral canal, a — bulb, b — rostellar sac, c — excretory **canal.** Mallory's phosphotungstic haematoxylin. (460×).
**Fig. 4.** Detail of the rostellar region in scolex with partly everted suckers. Musculature of **foreleg.** a — bulb, b — fibers of the rostellar sac, arrow — conically elongated apical part of **rostellum,** c — spiral canal. Mallory's phosphotungstic haematoxylin. (510×).


**Fig. 1.** Detail of the rostellar region in inverted scolex of *C. bovis* located in heart musculature. Note conspicuous praebulb (a) and bulb (b) extended into width and surrounded by fibers of the rostellar sac (c). Hooklets (arrow) visible in the tegument of right fold. Mallory's phosphotungstic haematoxylin. (1000 ×).

**Fig. 2.** Detail of the tegument of the left fold from Fig. 1 with well visible hooklets. Mallory's phosphotungstic haematoxylin. (1600 ×).

**Fig. 3.** Detail of the tegument of the right fold from Fig. 1 with well visible hooklets. Mallory's phosphotungstic haematoxylin. (1600 ×).

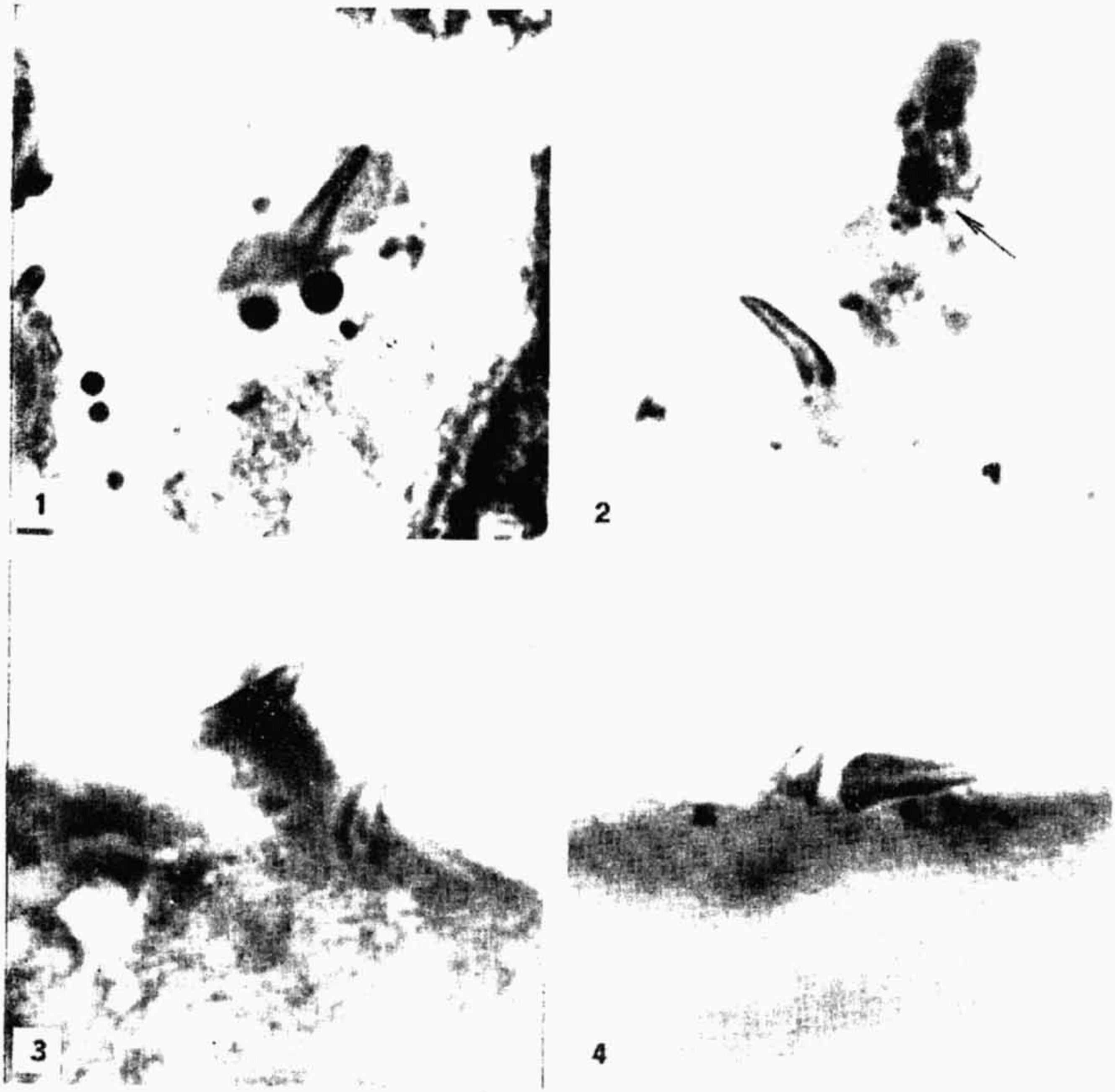


**Fig. 1.** Tangential section through the apical part of rostellum with well visible hook differentiated into cortical layer and medullar part. Mallory's phosphotungstic haematoxylin. (2000 ×).
**Fig. 2.** Cortical layer of hook and granules in the apical part of rostellum (arrow) are strongly positive to proteins with SS groups. PAA — aldehyde fuchsin. (2000 ×).
**Fig. 3.** and **Fig. 4.** Cortical layer of hooks contains a large amount of tyrosin. Morel Sisley's diazotization method. (2000 ×).