

## Morphometric Features of *Oesophagostomum dentatum*, *O. quadrispinulatum* and *Ascarops strongylina* in Materials from Wild Boars from Bulgaria

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A necropsy of wild boars from three regions of Southwest Bulgaria showed an infection with two oesophagostomid species (*Oesophagostomum dentatum* and *O. quadrispinulatum*) and one gastric spirurid (*Ascarops strongylina*). The collected parasites were used for a morphometric description of the species. The results were compared with the existing ones in the literature. It was established that the morphological features of the species from wild boars were the same as those from domestic pigs given by other authors. Diversity in the metric data was observed. However, metric features specific for the species were within the limits given by other authors.

*Key words:* *Oesophagostomum dentatum*, *Oesophagostomum quadrispinulatum*, *Ascarops strongylina*, wild boars, morphometric description

### Introduction

Pigs are even-toed ungulate animals from the family Suidae. They include the domestic pig and a number of other wild species, distributed all over the world. The European wild boar (*Sus scrofa scrofa* L.) is one of the most important game species in Europe, with populations that are increasing continuously [5]. The species can be found everywhere in Bulgaria and it is important from an ecological point of view. The wild boar is also important for the game-breeding and the hunting reserve. As omnivorous animals wild boars are susceptible to invasion by numerous species of parasites [18]. The most infected organs are lungs (with metastrongylids), stomach (with strongylids, spirurids), small intestines (with ascarids, acanthocephalids), caecum and colon (with trichurids, oesophagostomids), and liver (with trematodes).

Many authors in Bulgaria have studied the helminthofauna of the domestic pig [1, 2, 3, 6, 7, 9, 10, 20]. The studies for the wild boar are less [8, 11, 12, 14, 15]. During our recent studies in this aspect we have established two species of oesophagostomids (*Oesophagostomum dentatum*, *O. quadrispinulatum*) and one gastric spirurid (*Ascarops strongylina*) parasitizing wild boars. The available literature data on the morphology of these species are based only on materials from domestic pigs (*Sus scrofa domestica* L.) [3, 13, 16, 17, 19]. There is, however, no such data in the literature based on materials derived from wild boars. Therefore, the aim of the present study was to fill up this gap with providing morphometrical data on these three parasites in materials from wild boars spread on the territory of Bulgaria.

## Materials and Methods

The internal organs of 6 wild boars that were hunted during the shooting season in 2016 from three regions of Southwest Bulgaria were helminthologically necropsied. The discovered helminths were collected in a saline solution and after cleaning they were stored in 70% ethyl alcohol. Part of the helminths were enlightened in alcohol-glycerol sequence or with lactophenol, so that the taxonomic important features could be easier observed. After that the helminths were included in gelatin-glycerin and the edges of cover glasses were coated with Canadian balm for longer storage. The measurements were made with the help of classical parasitological methods or after shooting of separate structures of parasites with a Web camera Logitech 4000, which was attached to the “Amplival“ microscope, and their measuring with the picture analyzing computer program Image-Pro Plus-Version 6. Pictures were taken using a light microscope Leica DM5000 B, supplied with a camera and software (Leica Application Suite LAS v. 3.1). The obtained data were statistically analyzed according to Georgieva et al. [4]. Measurements in the tables are in  $\mu\text{m}$ .

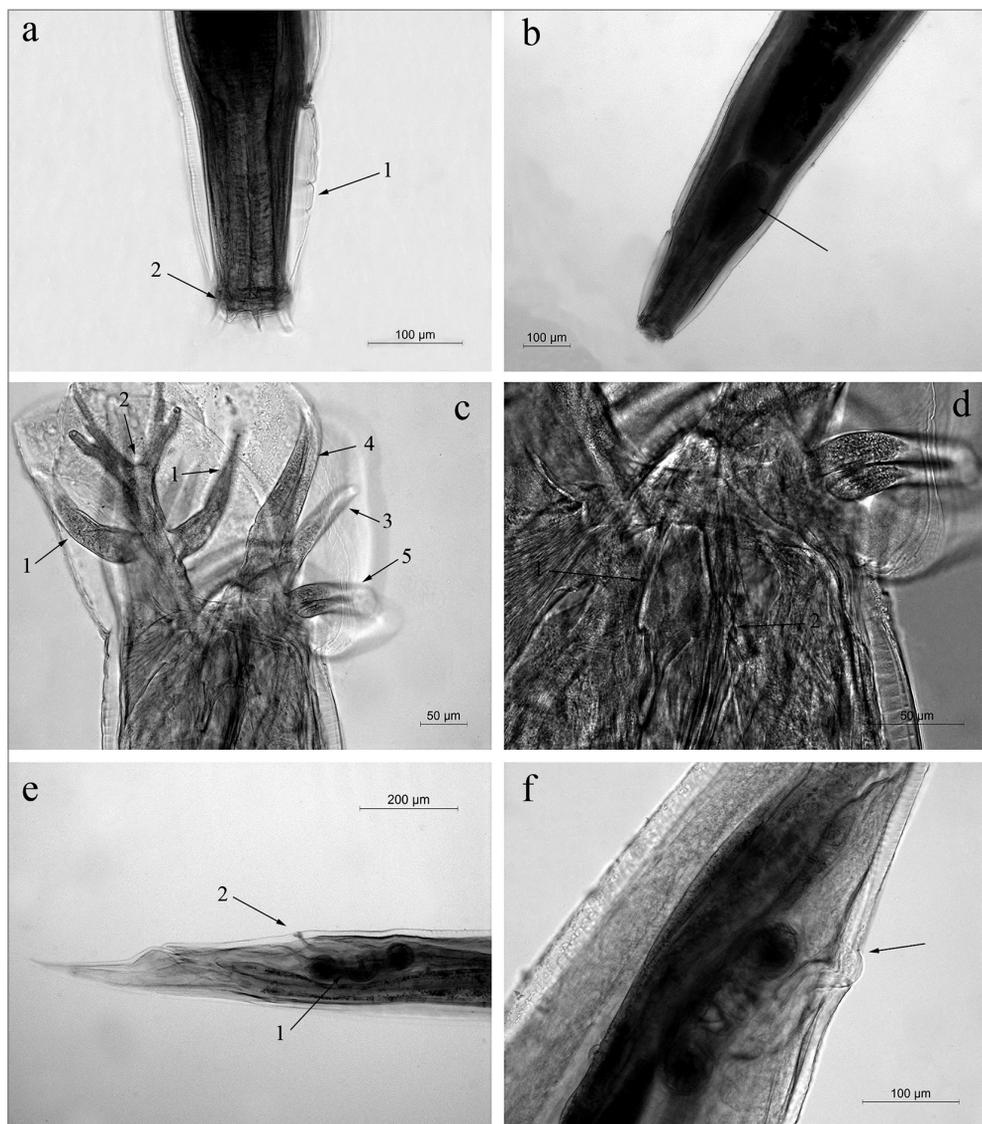
## Results and Discussion

We found two species of *Oesophagostomum* genus in the large intestines of two wild boars – *O. dentatum* and *O. quadrispinulatum*. In the stomach of one wild boar we found the gastric spirurid *Ascarops strongylina*. Further down data about their morphological and metrical features are supplied.

### ***Oesophagostomum* spp. – general features**

Oesophagostomids were white colored with a spindle shape. The two species had on their cranial end a vesicular cuticular dilatation. The dilatation begins from the buccal capsule and distally is restricted with a transversal cuticular line (**Fig. 1a**). The buccal capsule is formed around the mouth opening. It is relatively small, wider than longer and has two leaf crowns that consist of sharp lamellae (**Fig. 1a**). The outer crown is bigger and its sharp lamellae are leaf-shaped. The inner crown is poorly developed and has numerous small lamellae. The oesophagus begins after the buccal capsule, in its second half it is dilated and assumes a bulb-like form.

The male specimens have well-developed bursa copulatrix, genital cone, equally long spicules and spade-shaped gubernaculum which is pointed distally. The female oesophagostomids possess well-developed ovejector. The vulva opens in the distal end of the body, before anus.



**Fig. 1.** Morphologic features of *Oesophagostomum dentatum* in materials from wild boars from Bulgaria. a) anterior end: 1- vesicular cuticular dilatation, 2 - buccal capsule; b) oesophagus; c) bursa copulatrix: 1 - exterodorsal rays, 2 - dorsal ray, 3 - anterolateral ray, 4 - medio- and posterolateral rays, 5 - ventral rays; d) posterior end of male: 1 - gubernaculum, 2 - distal spicule end; e) Posterior end of female: 1- ovijector, 2 - vulva; f) opening of the vulva

### *O. dentatum* (Rudolphi, 1803), Molin, 186

The buccal capsule has a cylindrical form (**Fig. 1a**). The first half of the oesophagus has the same width and the second half is widened like a bulb (**Fig. 1b**). **Male.** The bursa copulatrix is massive with three well-developed parts (**Fig. 1c**). The dorsal ray and exterodorsal rays come from one wide trunk. First from the trunk are separated the exterodorsal rays. They do not reach the bursal edge. The trunk continues in the dorsal

**Table 1.** Metric data on *O. dentatum* by different authors

Structure	Ozerskaya 1930	Georgiev & Kamburov, 1977	Poelvoorde (1978)	Our data
Body length ♂ (in mm)	8.829	-	7.7 – 8.74	9.46± 0.28 (8.5-10)
Body width in the end of oesophagus ♂	-	-	-	218 ± 14 (184-255)
Oesophagus length ♂	-	-	-	397 ± 10.4 (364-428)
Max. oesophagus width ♂	-	-	-	110 ± 7,6 (81-128)
Spicule length	896-937	1030-1205	1004-1043	1135 ± 30 (1070-1250)
Gubernaculum length	101-122	92-138	-	127.7 ± 6.6 (111-148)
Body length ♀ (in mm)	7.5-13.4	-	8.72 - 10.83	11.5 ± 0.6 (9-12.4)
Body width in the end of oesophagus ♀	-	-	-	233.5 ± 14 (182-260)
Oesophagus length ♀	-	-	-	426.6 ± 10.2 (406-451)
Max. oesophagus width ♀	-	-	-	115.7 ± 5.8 (101-126)
Vulva - anus	315-366	265-393	-	347 ± 15.8 (280-370)
Anus - tail tip	255-265	241-324	313	293 ± 7.4 (270-310)

ray, which later is divided into two branches. At a half of their length those branches are also divided into one shorter lateral branch and one longer medial branch. Only the medial ones reach the bursal edge. The lateral rays also come out from one trunk. The anterolateral ray is the first separated from the trunk and it is the shortest from the lateral rays. The other two lateral rays are differentiated a little after that and stay adhering to each other until their end. The mediolateral ray is a little shorter than the posterolateral one. The ventral rays have the same length and also begin from one trunk. These rays differentiated early but stay stuck together for all of their length. The spicules are equal in length. Their proximal end is round and thickened and distal end is sharpened (**Fig. 1d**). The gubernaculum looks like a spade directed distally (**Fig. 1d**). **Female.** The tail has a form of prolonged cone (**Fig. 1e**). The ovejector is chitinized and composed of two round parts and a channel system, which passes into the vulva (**Fig. 1e, f**). The metric data for this species in our materials together with the measurements from other authors that were available in the literature are shown in **Table 1**.

When we have compared our data from those by Ozerskaya [16] and Poelvoorde [17] who have used materials from domestic pigs, it can be seen that our male specimens as a whole are bigger. The mean values for the body length, spicules and gubernaculum exceed the maximal values pointed by those authors. The same is assigned to the distance between the anus and tail tip in females, measured by Ozerskaya [16]. However, Poelvoorde [17] has measured a longer distance between the anus and tail tip

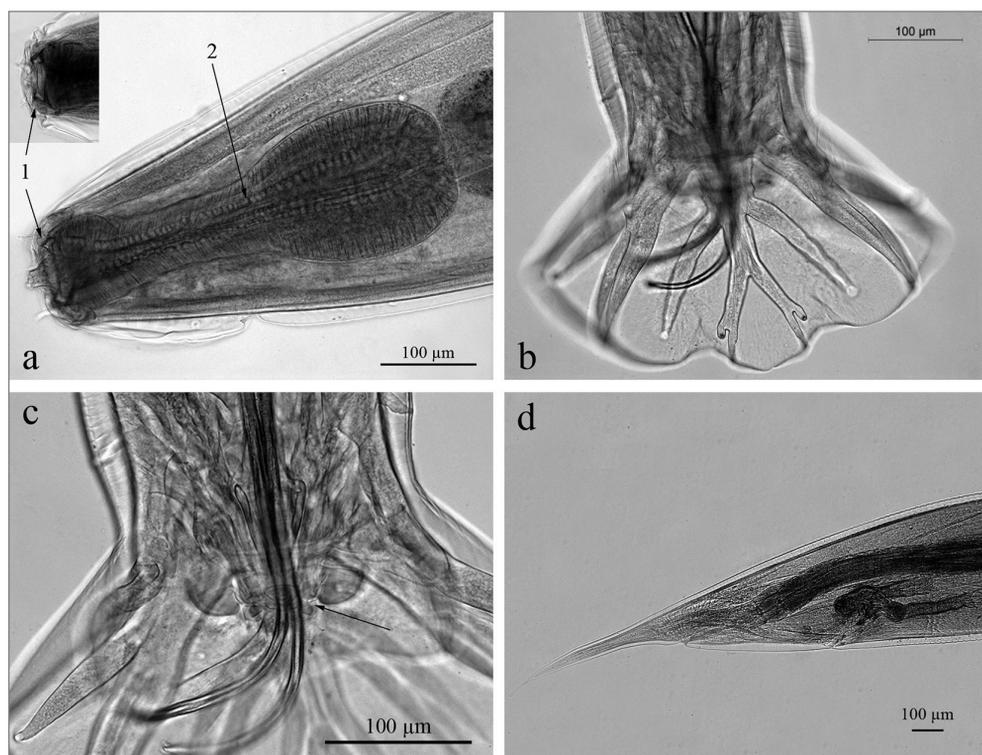
than us. Our metric data for this species are completely overlapped with those pointed by Georgiev and Kamburov [3] in materials from domestic pigs from four regions of Bulgaria, including the Sofia region.

### ***O. quadrispinulatum* Marcone, 1901**

The buccal capsule (**Fig. 2a**) has a form of a truncated cone with the basis directed at the oesophagus. The oesophagus (**Fig. 2a**) has two dilatations: one small round widening at the beginning and a bulbous widening in its second half. The oesophagus canal is surrounded by a row of round light-refracting cells.

**Male.** The bursa copulatrix, spicules and gubernaculum have the same morphology as *O. dentatum* (**Fig. 2 b, c**). The genital cone is well-developed and there are some papillae on its surface (**Fig. 2c**). **Female.** The tail is thin and styloform. The ovejector is similar to this of *O. dentatum* (**Fig. 2d**). Our metric data and the available literature data on this species are shown in **Table 2**.

In Bulgaria *O. quadrispinulatum* has been reported by Georgiev and Kamburov [3]. They have found it in domestic pigs from 4 regions, including the Sofia region, where our materials from wild boars originated too. The metric data from those authors almost entirely corresponds with our data. There is a lack of correspondence only about the length of females. It is about 1 mm higher than that from our materials. Our data also



**Fig. 2.** Morphologic features of *Oesophagostomum quadrispinulatum* in materials from wild boars from Bulgaria. a) Anterior end: 1- buccal capsule, 2 - oesophagus; b) posterior end of male; c) genital cone; d) posterior end of female

**Table 2.** Metric data on *O. quadrispinulatum* by different authors

Structure	Mapelstone, 1930	Georgiev & Kamburov, 1977	Our data
Body length ♂ (in mm)	6.56-8.85	7.8-8.8	8.13 ± 0.53 (6.8-9)
Body width in the end of oesophagus ♂	–	–	217 ± 15.7 (180-243)
Oesophagus length ♂	–	380-420	402 ± 13 (371-420)
Max. oesophagus width ♂	–	–	115 ± 7.7 (105-125)
Spicule length	780-880	765-955	843 ± 63 (760-990)
Gubernaculum length	104-120	85-128	105.6 ± 5.6 (93-115)
Body length ♀ (in mm)	8.33-10.36	9.6 - 12.1	9.93 ± 0.62 (8-11.1)
Body width in the end of oesophagus ♀	–	–	266.8 ± 24.8 (210-330)
Oesophagus length ♀	–	426 - 462	439.8 ± 8 (416-456)
Max. oesophagus width ♀	–	–	133 ± 5.6 (118-156)
Vulva – anus	380-520	400 - 552	445.4 ± 19.2 (400-500)
Anus – tail tip	360-370	458 - 588	530.1 ± 22 (460-600)

correspond to those pointed by Mapelstone [13] in materials from domestic swine from Bengal. There is only one exception concerning the distance between the anus and tail tip in females, which is longer in our specimens.

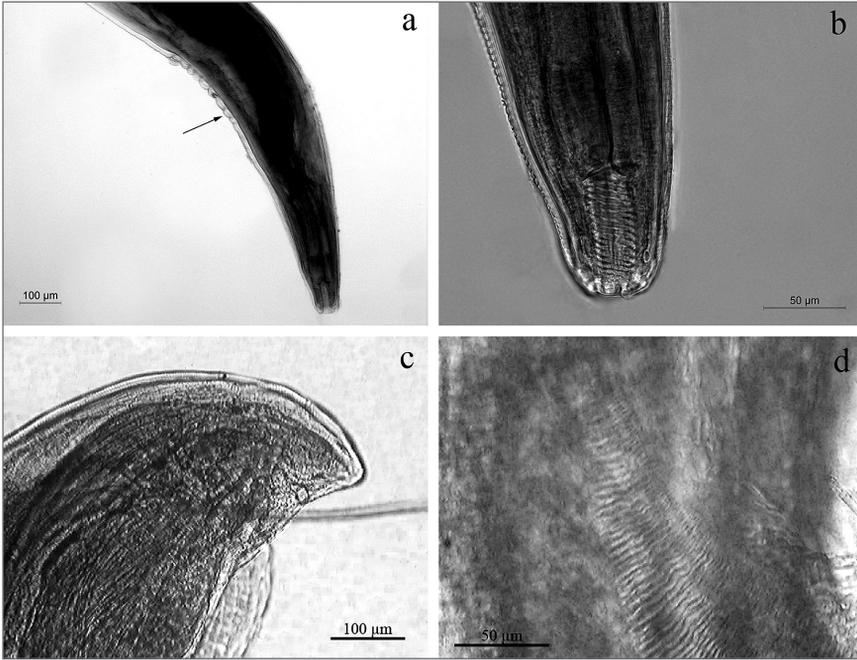
The two oesophagostomid species are distinguished by some metric and morphological features. *O. dentatum* has a longer body than *O. quadrispinulatum*. The spicules of *O. dentatum* are longer too. The proportion between the thin and leaf-shaped parts of the gubernaculum in *O. dentatum* is 1:1. In *O. quadrispinulatum* this ratio is 1:2. *O. quadrispinulatum* specimens have longer distances between the vulva and anus and between the anus and tail tip than these of *O. dentatum*. The oesophagus of *O. dentatum* usually has not light-refracting structures or these structures can be found only in its beginning. The oesophagus of *O. quadrispinulatum* has a small, but clearly visible second dilatation right after the buccal capsule.

### *Ascarops strongylina* Rudolphi, 1819

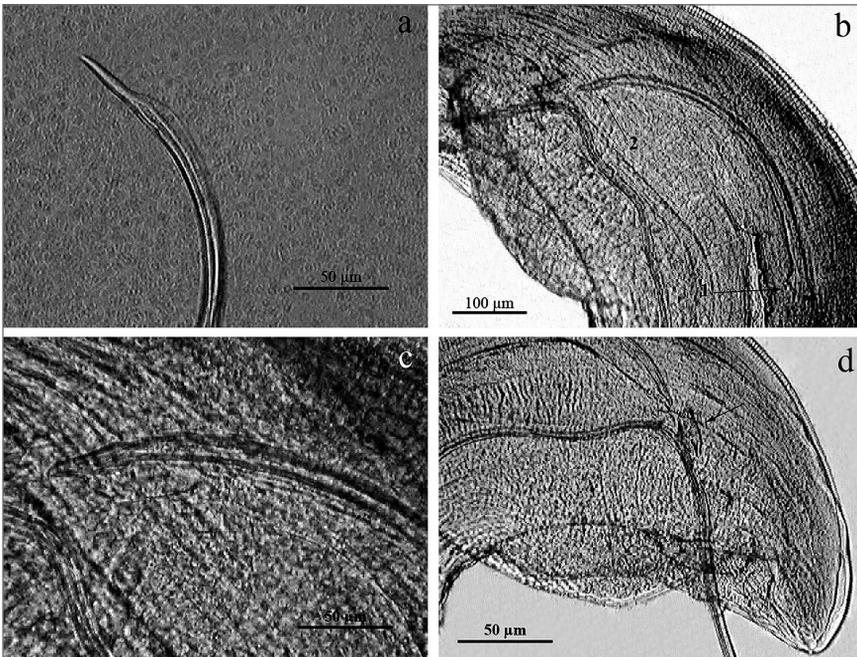
This is a brown nematode with fusiform form. It has one narrow lateral cuticular wing (Fig. 3a). From the mouth opening begins the pharynx, which is consisted of spiral rings (Fig. 3b). The oesophagus has two parts – the anterior part is muscular and short and posterior is longer and glandular.

**Male.** There is no bursa copulatrix. The posterior end is curved ventrally. Two asymmetric, transversally furrowed wings are visible on it, as one is wider than another (Fig. 3c, d). Around the cloaca are several pairs of genital papillae. The spicules are different in length and form. The one spicule is long and thin. Its distal end is lancet-shaped (Fig. 4a).

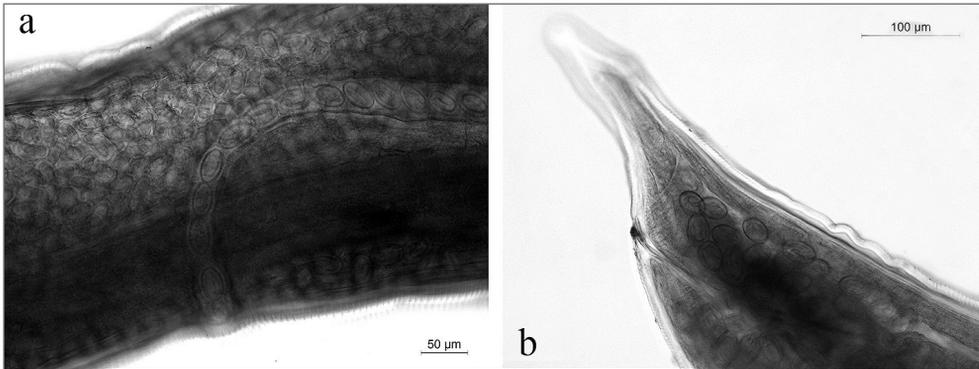
The second spicule is shorter and wider, its proximal end is thickened and distal end looks like an arrow-head (Fig. 4b, c). The gubernaculum is situated around the cloaca and looks like an elongated lamella (Fig. 4d). **Female.** The vagina is tubular and



**Fig. 3.** Morphologic features of *Ascarops strongylina* in materials from wild boars from Bulgaria. a) anterior end – lateral cuticular wing; b) anterior end – pharynx; c) posterior end of male; d) cuticular wing



**Fig. 4.** Morphologic features of *Ascarops strongylina* in materials from wild boars from Bulgaria. a) distal end of big spicule; b) small spicule: 1 – proximal end, 2 – distal end; c) distal end of small spicule; d) gubernaculum



**Fig. 5.** Morphologic features of *Ascarops strongylina* in materials from wild boars from Bulgaria. a) vagina and vulva; b) posterior end of female

**Table 3.** Metric data on *Ascarops strongylina* by different authors

Structure	Shmytova, 1963	Our data
Body length ♂ (in mm)	10.1-14.9	12
Body width in the end of oesophagus ♂	270 - 360	330
Anterior end - beginning of lateral wing ♂	210 - 290	211
Length of the pharynx ♂	80-100	86
Width of the pharynx ♂	30	32
Number of pharynx's spiral rings in ♂	14-18	17
Length of muscular oesophagus ♂	300-400	350
Length of glandular oesophagus ♂	2340-3700	3200
Oesophagus width ♂	-	119
Small spicule length	500-600	575
Big spicule length	2300-2900	2100
Gubernaculum length	66-80	55
Gubernaculum width	50-69	38
Width of the bigger caudal wing ♂	200-230	290
Cloaca – tail tip	-	253
Body length ♀ (in mm)	12.4-23.7	16.52 ± 2.76 (14-20)
Body width in the end of oesophagus ♀	230 - 430	357.8 ± 37.9 (300-382)
Anterior end - beginning of lateral wing ♀	230 - 340	340 ± 67 (288-420)
Length of the pharynx ♀	90-105	83 ± 8.2 (74-92)
Width of the pharynx ♀	30-40	31 ± 1.8 (29-33)
Number of pharynx's spiral rings in ♀	14-18	15-16
Length of muscular oesophagus ♀	290-540	359 ± 12.1 (345-370)
Length of glandular oesophagus ♀	2730-3800	3360 ± 400 (2900-3800)
Oesophagus width ♀	-	124 ± 13.6 (108-137)
Anterior length - vulva	6800-1110	8120 ± 1340 (6800-9400)
Anus - tail tip	230-300	232 ± 5.9 (219-240)

situated in the middle of the body. At first it goes parallel to the body, afterwards turns perpendicular and opens in the vulva (**Fig. 5a**). The eggs in the vagina are embryonated. The body's caudal end is straight and the tail is short with a rounded tip (**Fig. 5b**).

We compared our metric data for *A. strongylina* with those from description by Shmytova [19]. Comparative data are shown in **Table 3**.

Our results about most of the measurements correspond with hers. There are few differences in the females: the lateral cuticular wing of specimens from our materials starts backwards, the pharynx is shorter and its width is in the lower limits pointed by Shmytova [19]. More important differences are observed in the male sexual structures. Our male specimen has shorter big spicule and gubernaculum, whereas its bigger cuticular wing on the caudal end is wider.

## Conclusion

The morphological features of the established by us species *O. dentatum*, *O. quadrispinulatum* and *A. strongylina* in wild boars from Bulgaria completely correspond to those described by other authors in materials from domestic pigs. Diversity in the metric features is observed, which could be due to differences in methods of preparation and observation of the helminths as well as to the peculiarities of the separate parasite populations from different parts of the world. The measurements about taxonomic important features, however, are in the limits, which have been pointed by other authors.

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