

PARASITOLOGY

ПАРАЗИТОЛОГИЯ



УДК 576.895.132:616.995.132

Original Empirical Research

<https://doi.org/10.23947/2949-4826-2025-24-3-7-13>

Trichinellosis in Badgers in the Rostov Region (Southern Russia)

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EDN: VQIJZO

Abstract.

Introduction. Nematodes of the genus *Trichinella* are the causative agents of trichinellosis, which affects humans and animals and causes significant economic damage to agricultural and food industries, especially with regard to food safety. Currently, the genus *Trichinella* includes 10 species and 3 genotypes, which are divided into 2 clades: encapsulated and non-encapsulated. In the wildlife, a wide range of animals participate as carriers in circulation of *Trichinella*, and badgers are one of the main natural reservoirs for this disease. The article provides the information on detection of the first case of *Trichinella* larvae in a badger in the Rostov region.

Materials and Methods. The object of the study was a carcass of a badger hunted in the Sholokhov district of the Rostov region in August 2024. Detection with further study of larvae and capsules in the samples was carried out by the method of compressor trichinelloscopy. Afterwards, histological sections were made from the individual muscles, which were stained with hematoxylin and eosin according to the standard technique.

Results. Capsules of *Trichinella* were found in all the examined muscles of an animal. The largest number of them was found in the diaphragm and its crura. The larvae found belonged to the encapsulated ones and were located in capsules in groups or one at a time. Histological sections showed thick collagen shells of the capsules surrounded from the outside by thin membranes of connective tissue. The results of capsule morphometry showed that their size didn't differ depending on their location in the badger's body; the shape of the capsules in all muscles was almost round.

Discussion and Conclusion. The capsule shape indices calculated based on the morphometric results are closer to the corresponding indices of *T. nativa* than *T. spiralis*, however, this does not allow us to make fully accurate conclusions about the species membership of *Trichinella*, since the molecular genetic studies or the use of Western blot are required for precise diagnostics. Thus, the study of *Trichinella* species distribution and composition remains an extremely relevant objective that requires further research both in our region and worldwide.

Keywords: *Trichinella*, *Meles meles*, trichinellosis, morphometry, larvae, capsules

Acknowledgements. The authors are grateful to the hunters who provided the carcass of a badger for the study. The authors are also grateful to M.S. Kaple (DSTU) for preparing histological sections.

For Citation. Evsyukov AP, Potapenko IO, Zelenkov AP, Zelenkova GA. Trichinellosis in Badgers in the Rostov Region (Southern Russia). *Russian Journal of Veterinary Pathology*. 2025;24(3):7–13. <https://doi.org/10.23947/2949-4826-2025-24-3-7-13>

Трихинеллез у барсука в Ростовской области (юг России)

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Аннотация

Введение. Нематоды рода *Trichinella* — возбудители трихинеллеза, поражающего людей и животных и наносящего значительный экономический ущерб сельскому хозяйству и пищевой промышленности, особенно в сфере безопасности продуктов питания. В настоящее время род *Trichinella* включает 10 видов и 3 генотипа, которые разделяются на 2 кланды: капсулообразующие и бескапсульные. Циркуляция трихинелл в дикой природе происходит при участии обширного круга животных-трихинеллоносителей, и барсук является одним из основных природных резервуаров заболевания. В статье приводятся сведения о первом случае обнаружения личинок трихинелл у барсука в Ростовской области.

Материалы и методы. Объектом исследования послужила туша барсука, добытая охотниками в Шолоховском районе Ростовской области в августе 2024 г. Выявление и изучение личинок и капсул в образцах проводили методом компрессорной трихинеллоскопии. В дальнейшем из отдельных мышц были изготовлены гистологические срезы, которые окрашивались гематоксилином и эозином по стандартной методике.

Результаты исследования. Капсулы трихинелл были обнаружены во всех исследованных мышцах животного. Наибольшее их количество найдено в диафрагме и ее ножках. Обнаруженные личинки относились к инкапсулирующим и располагались в капсулах группами или по одной. На гистологических срезах капсулы имели толстые коллагеновые стенки, окруженные снаружи тонкими соединительнотканными оболочками. Результаты морфометрии капсул показали, что они не отличаются по размерам в зависимости от локализации в теле барсука, форма капсул во всех мышцах была почти круглой.

Обсуждение и заключение. Индексы формы капсул, рассчитанные по результатам морфометрии, более близки к соответствующим показателям *T. nativa*, чем *T. spiralis*, однако это не позволяет делать точные выводы относительно видовой принадлежности трихинелл, поскольку для точной диагностики необходимы молекулярно-генетические исследования или использование вестерн-блота. Таким образом, изучение распространения и видового состава трихинелл по-прежнему остается крайне актуальной задачей, требующей дальнейших исследований как в нашем регионе, так и по всему миру.

Ключевые слова: *Trichinella*, *Meles meles*, трихинеллез, морфометрия, личинки, капсулы

Благодарности. Авторы статьи выражают благодарность охотникам, предоставившим для исследования тушу барсука. Также авторы признательны М.С. Капле (ДГТУ) за изготовление гистологических срезов.

Для цитирования. Евсюков А.П., Потапенко И.О., Зеленков А.П., Зеленкова Г.А. Трихинеллез у барсука в Ростовской области (юг России). *Ветеринарная патология*. 2025;24(3):7–13. <https://doi.org/10.23947/2949-4826-2025-24-3-7-13>

Introduction. Nematodes of the genus *Trichinella* of the family Trichinellidae, are the causative agents of trichinellosis, the intestinal and tissue type of helminthiasis deemed to be one of the most serious and widespread zoonotic infections threatening human health [1]. In humans, this disease has been reported in 55 countries worldwide [2]. Apart from its negative impact on public health, trichinellosis causes

significant economic damage to agricultural and food industries, particularly with regard to food safety.

Trichinella have a simple yet unique life cycle. The enteric phase begins with the consumption of meat containing tissue parasite capsules. Larvae are released from the capsules due to gastric acid activity and, within a few hours, migrate to the small intestine, where they penetrate into the

columnar epithelium. There, they undergo four molts to mature and begin mating 30 hours after infestation. On the fourth day, newborn larvae migrate through the blood-vascular system to the striated muscles and penetrate into muscle cells. Here, encapsulated species form collagen capsules and develop into an invasive form after 15–20 days [3]. Larvae of non-encapsulated species are likely to migrate between muscle layers [4]. Thus, a characteristic feature of the *Trichinella* life cycle can be identified: any animal they parasitize in becomes for them both a definitive and intermediate host.

At present, the genus *Trichinella* includes 10 species and 3 genotypes having no taxonomic status. All known species and genotypes are clearly divided into 2 clades. One clade includes *Trichinella* that encapsulate in the muscle tissue of a host and are capable of infecting mammals. It includes *T. spiralis*, *T. nativa*, *T. britovi*, *T. murrelli*, *T. nelsoni*, *T. patagoniensis*, *T. chanchalensis* and genotypes designated as T6, T8 and T9. The second clade includes three species of *Trichinella* that do not encapsulate: among them, one species infests mammals and birds (*T. pseudospiralis*); and two species parasitize in mammals and reptiles (*T. papuae* and *T. zimbabwensis*) [3, 5]. All of the above-mentioned species and genotypes of *Trichinella* pose danger to both animals and humans. Primarily one species was previously reported in the Rostov Region: *T. spiralis* [e.g., 6]. In the Krasnodar region, cases of finding the non-encapsulated species *T. pseudospiralis* in pigs have been recorded [7].

In the wildlife, a wide range of animals participate as carriers in circulation of *Trichinella*, which includes approximately 100 diverse species of carnivorous and omnivorous animals [8]. A badger (*Meles meles*) is one of the main natural reservoirs of trichinellosis [9]. In the Rostov Region, as well as in the whole country, badgers are often the main prey for hunters. According to data published on the website of the Ministry of Natural Resources and Environment in July 2024¹, the population of badgers in the Rostov Region equals to 2 556 specimens, and the authorised quota for hunting is 3.91% of the population. Although this is quite a high level, poaching is

still taking place. As a result, the uninspected meat can be consumed by humans or animals, which can lead to their infection with trichinellosis.

In the article, we report the first case of detecting *Trichinella* larvae in a badger in the Rostov Region.

Materials and Methods. The object of the study was a carcass of a badger hunted on the territory of the Veshensky Production and Experimental Hunting Area (Sholokhov District, northern part of Rostov Region) in August 2024. Various muscle groups were sampled, including the crura of the diaphragm, diaphragm, intercostal muscles, neck muscles, and thigh muscles. Larvae and capsules in these samples were isolated and examined using compressor trichinoscopy. Histological sections were then prepared from separate muscles and stained with hematoxylin and eosin according to the standard techniques. Capsules in native samples were measured and photographed using a Levenhuk MED D45 microscope (Levenhuk Inc., China) and LevenhukLite software. The capsule shape index (V) was calculated as the ratio of the capsule length (L) to its diameter (D). This index is used to evaluate the shape of round objects, including *Trichinella* capsules [10, 11].

Results. *Trichinella* capsules were found in all the examined muscles of an animal. The largest number was found in the diaphragm and its crura. It has been acknowledged in the previous studies that the most heavily infested muscles in badgers are the head muscles, particularly the mylohyoid muscle [12]. However, these muscles were not within the scope of the present research.

The larvae discovered belong to the encapsulated ones. They were located in capsules in groups or one at a time (Fig. 1 a–b). Histological sections revealed that the capsules had thick shells covered by thin connective tissue membranes formed by the organism of a host (Fig. 1 c–d).

Based on the capsule morphometric data obtained (Table 1), it is possible to state that capsule location in the badger's body does not affect its size to any significant extent. The shape of capsules in all the muscles was nearly round ($V \approx 0.79$).

¹ Directive of the Governor of the Rostov Region of July 17, 2024 No. 175 “On approval of quota for obtaining hunting resources in the Rostov Region for the period from August 1, 2024 to August 1, 2025”. URL: <https://минприродыро.рф/upload/uf/5b8/n4ub6dzirqhmt4tivn963j39r4zpdks/limity.pdf> (accessed: 05.06.2025 r.).

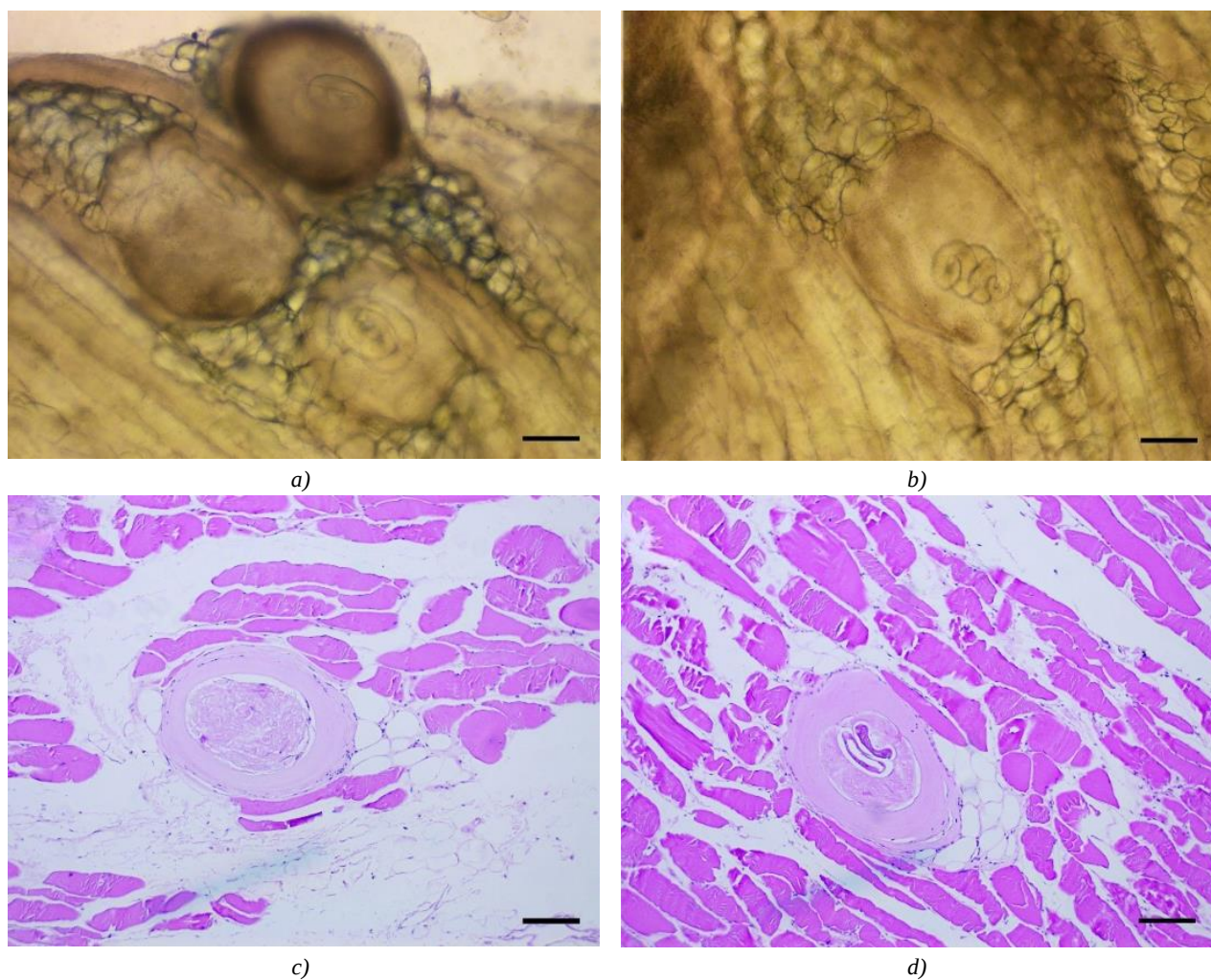


Fig. 1. *Trichinella* capsules with larvae in muscles of a badger: *a-b* — native preparation; *c-d* — histological sections stained with hematoxylin and eosin. Scale: 0.1 mm

Table 1

Morphometric parameters of *Trichinella* capsules in various muscles of a badger

Muscles examined	Number of capsules	Capsule diameter (D), mm	Capsule length (L), mm	Shape index (V)
Crura of the diaphragm	20	0,441±0,010	0,349±0,008	0,797±0,020
Diaphragm	20	0,438±0,007	0,342±0,020	0,791±0,010
Intercostal muscles	20	0,440±0,009	0,348±0,010	0,790±0,008
Neck muscles	20	0,438±0,011	0,349±0,015	0,798±0,012
Thigh muscles	10	0,437±0,020	0,350±0,011	0,801±0,020

Discussion and Conclusion. The capsule shape indices calculated based on the morphometric results correspond more to the respective indices of *T. nativa* rather than *T. spiralis* [11]. The former species used to be registered in the Central Russia, but has not been previously observed in the south of the country or in the Caucasus [13]. The -4°C isotherm in January is likely to be a southern boundary for this species [4], which makes the possibility of finding this species in the Rostov Region very low. However, it is impossible to draw precise conclusions regarding the species membership of *Trichinella* detected by us in the badger, since the molecular genetic studies [14] or the use of Western blot technique [15] are required for a reliable diagnosis. Moreover, it is worth noting that most *Trichinella* species have a sympatric distribution [3]. Thus, in the Rostov region there could potentially be detected two encapsulated species (*T. spiralis* — reported here before and distributed

worldwide, and *T. britovi* — disseminated from the Iberian Peninsula to Kazakhstan, Turkiye and Iran [4]), and one non-encapsulated species — *T. pseudospiralis* registered in North America, Eurasia and Tasmania [16]. No evident dependence of different *Trichinella* species to host animals has been revealed [17], however, different invasion rate by different *Trichinella* species have been recorded during experimental infestation [18]. All the above makes species identification of *Trichinella* extremely difficult.

Thus, studying the distribution and species composition of *Trichinella* remains a highly relevant objective, requiring further research both in south of Russia and worldwide. Badger meat must undergo mandatory trichinosis to prevent potential infestations in humans. Unused carcasses and their remains must be disposed of to exclude infestation of domestic and wild animals.

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Claimed Contributorship:

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Conflict of Interest Statement: the authors declare no conflict of interest.

All authors have read and approved the final manuscript.

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Заявленный вклад авторов:

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И.О. Потапенко: поиск литературы, подготовка текста, формирование выводов.

А.П. Зеленков: цели и задачи исследования, подготовка текста, формирование выводов.

Г.А. Зеленкова: цели и задачи исследования, подготовка текста, формирование выводов.

Конфликт интересов: автор заявляет об отсутствии конфликта интересов.

Автор прочитал и одобрил окончательный вариант рукописи.

Received / Поступила в редакцию 28.05.2025

Reviewed / Поступила после рецензирования 25.06.2025

Accepted / Принята к публикации 30.06.2025