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RESEARCH

First evidence of *Brachylaima* sp. (Diplostomida; Brachylaimidae) infesting the Etruscan shrew (*Suncus etruscus*) in Iran

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Abstract

Of the eleven species of shrews described in Iran, the Etruscan shrew (*Suncus etruscus*), the smallest known extant mammal by mass, is recorded in northern and southern provinces. During rodent control programs, a female *S. etruscus* was trapped and found naturally infected with a trematode which was morphologically identified as *Brachylaima* sp. This finding represents the first report of trematodiasis in this host species.

Keywords *Brachylaima* · *Suncus etruscus* · Trematodiasis · Iran · Case report

Introduction

Shrews (family Soricidae) are small mammals belonging to the order Eulipotyphla (Wilson and Reeder 2005). Of the eleven species presently described in Iran (Etemad 1984), the genus *Suncus* is widely distributed throughout the country, though its natural history and ecology are not well known (Etemad 1984). The Etruscan shrew (*Suncus etruscus*) is widely distributed from Europe and North Africa up to South Asia and some Mediterranean islands (Jürgens 2002), as well as in the northern and southern provinces of Iran (Karimii et al. 2008; Mahmoudi et al. 2017). They appear brownish-gray, with small body size (i.e., from 40 to 56 mm and a weight of 1.8–3 g) (Mahmoudi et al. 2017). They are agile creatures with a fast metabolism, which imposes a high daily food consumption of about 2 times their body weight (Hanski 1984). Their diet includes diverse types of invertebrates like snails, earthworms, insects, as well as tiny lizards and newborn rodents (Etemad 1984). Due to the wide range of their eating preferences, they harbor a plethora of parasites while ingesting particular infected intermediate hosts (Hall 1929), including *Brachylaima* spp. (Butcher 2016). Trematodes of the Brachylaimidae family consist of seven genera and over seventy species of endoparasites infesting vertebrates, including mammals, birds, reptiles, and amphibians as definitive hosts (Sirgel et al. 2012). These helminths have a global distribution, being occasionally reported in human patients in Australia (Butcher 2016; Butcher et al. 1998). These trematodes have a terrestrial life cycle that includes land snails and slugs as first and/or second intermediate hosts (Butcher 2016). While zoonotic helminths of small mammals have been documented in Iran (Mohtasebi et al. 2020), data about shrews is scant. In this study, we reported *Brachylaima* sp. in *S. etruscus* in Southwestern Iran, with emphasis on the possible role that this small mammal could play in the ecology of this trematode.

Case report

Study area

The study was conducted on the southwest bank of the Karun River in Ahvaz City (31.254019, 48.583810; Fig. 1).

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Fig. 1 Study area, Ahwaz City, Khuzestan Province, Iran

Ahwaz City has a unique subtropical climate, and the Karun River is the largest and most water-rich river in Iran, with an approximate length of 900 km, dividing Ahvaz City into eastern and western halves.

Sample collection and identification

During an authorized rodent control program that was done under the Ahvaz City municipality regulations in the late 1990s, a female *S. etruscus* was accidentally trapped in a live trap. Due to delayed trap collection, the shrew died, and the carcass was transferred to the Ahwaz Health Research Station affiliated with the School of Public Health at Tehran University of Medical Sciences. The identification of the shrew species was performed based on morphological and morphometric measures using key references (Etemad 1984).

Isolation of parasitic helminths from rodent intestines and identification

Dissection of the preserved sample in formalin was carried out at the Helminthology Laboratory of the School of Public Health years later, aiming to reveal the parasite. The organs were placed in tap water in Petri dishes and were precisely examined for the presence of helminth under a stereomicroscope. The only detected helminth found in the shrew was stained with carmine stain and mounted using Canada balsam. The identification of the isolated helminth as *Brachylaima* sp. was conducted based on taxonomic keys (Yamaguti 1958), and morphological characteristics were described by drawings with a camera lucida-equipped microscope. Moreover, the entire intestinal contents were microscopically examined for any parasite's ova. Measurements of the recovered trematode are as follows: length of the body was 3.15 mm; width of the body was 0.80 mm; oral sucker 295 × 280 µm; acetabulum 289 × 311 µm; pharynx 187 × 163 µm; ovary 220 × 233 µm; the size of the anterior testicle was 280 × 360 µm; the size of the posterior testicle was 308 × 315 µm; the size of eggs were 32–35 × 17–20 µm. It should be noted that in the current case, the esophagus is absent and the uterus does not surpass the intestinal caeca, neither bilaterally nor anteriorly, and the vitelline glands only extend up to the posterior level of the acetabulum as depicted (Fig. 2).

Discussion

The present paper illustrates the first natural occurrence of *Brachylaima* sp. trematode in an Etruscan shrew. In 1843, Dujardin described *Brachylaima advena* as a new genus and species of a trematode in a bicolored white-toothed shrew (*Crocidura leucodon*), identified on the verge of Rennes, France (Dujardin 1843). Complementary studies on the life-cycle, experimental infections as well as other infected hosts, confirmed the speculation of Dujardin (Joyeux and Foley 1930). Numerous experimental studies have been conducted to determine the lifecycle of some Brachylaimid helminths and their definitive and intermediate hosts. In similar laboratory studies, the life cycles of *B. cribbi* and *B. aspersae* were elucidated with an emphasis on the developmental stages of the parasite's intermediate hosts as well as precise morphological descriptions (Butcher and Grove 2001; Segade et al. 2011).

Brachylaima spp. has been reported from the insectivores *Crocidura russula* and rodents *Eliomys quercinus* in the Serra Calderona mountains, Valencian Community, Spain (Fuentes

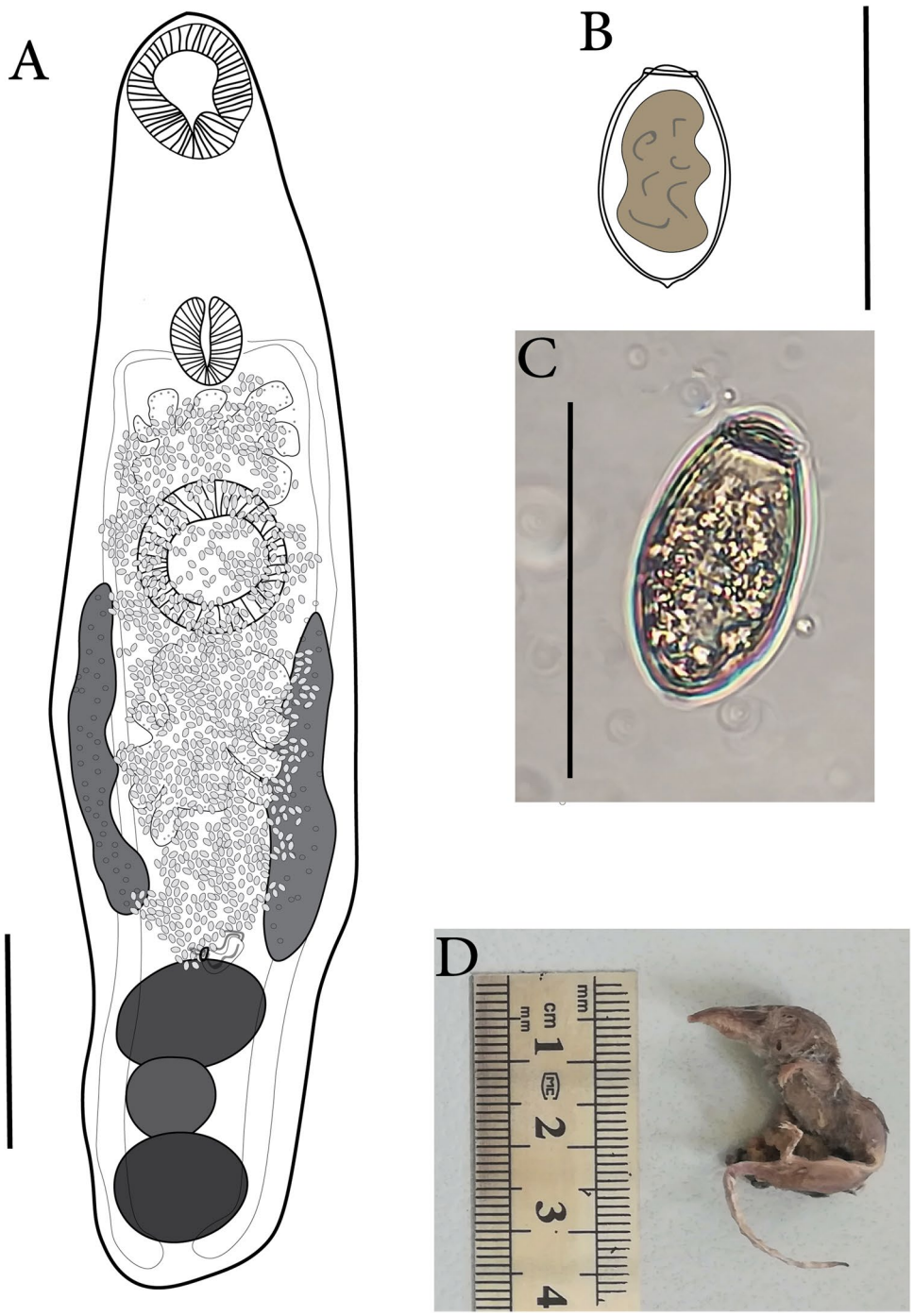

Fig. 2 **A** *Brachylaima* sp. drawn by camera lucida, bar: 500 µm. **B** and **C** *Brachylaima* sp egg, bar: 40 µm. **D** *S. etruscus*, the host of *Brachylaima* sp

et al. 2000). *Brachylaima* spp. was detected in the wood mouse (*Apodemus sylvaticus*) in Serra Calderona Natural Park (Valencian Community, Spain) (Debenedetti López 2015). Brachylaimid specimens were also discovered in the small intestines of a male and a female greyhound coming from a village located in a valley in the North of Spain (Guisantes et al. 1994). In Iran, few studies have been conducted on the presence of Brachylaimids, and in one study, *Brachylaima* sp. was reported from a stray dog in north Iran (Mobedi et al. 2017), and another study has described this trematode in a dog in North Khorasan province (Heidari 2016). Neither study identified the parasite to species level. Although numerous zoological studies have been conducted on shrews in Iran, parasitological studies on them are rare. In one study on *C. leucodon* in Dasht-e-Razan, Western Iran, 9 species of endoparasites and ectoparasites were reported, none of which were trematodes (Yousefi et al. 2017). However, in terms of the parasitic fauna of *S. etruscus*, as the smallest mammal in

the world, no data had been presented until the 1980s, when infections with three new species of cestodes of *Hymenolepis claudevaucheri*, *H. cerberensis*, and *H. banyulsensis* were reported, and later, reallocated to the genus *Staphylocystis* belonging to the family Hymenolepididae (Mas-Coma et al. 1984, 1986a, 1986b). Recently, a unique study on a significant sample size of 166 individuals of *S. etruscus* in France revealed a parasitic infection rate of 50.6% in the captured shrews and *Joyeuxiella pasqualei* larvae, *Mesocestoides* sp. larvae, *S. claudevaucheri*, *S. banyulsensis*, *S. cerberensis*, and *Pseudhymenolepis* sp. and the nematode *Aonchotheca* sp. were identified (Galán-Puchades et al. 2021).

From a public health point of view, human infections with *B. cribbi* have been described in two children (Butcher et al. 1996) and an adult (Butcher et al. 1998) from South Australia, all presenting symptoms such as mucoid, watery diarrhea, abdominal pain, and anorexia. In these patients, treatment with praziquantel was successful (Butcher et al. 1996), further suggesting the parasitic origin of the clinical presentation.

In terms of the limitations of the current study, as only one trematode was recovered and was preserved in formalin, it was not possible to conduct molecular studies. Additionally, owing to the fact that *Brachylaima* spp. are morphologically similar and their lifecycle is unknown, the parasite was only identified at the genus level. In conclusion, the present study is the first report of trematode infection in *S. etruscus*, the smallest living mammal in the world. Molecular studies to elucidate parasite diversity and host-parasite interaction in *S. etruscus* should be performed, and the possible role of shrews in spreading zoonotic pathogens should be investigated.

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Author contribution FN drafted the manuscript and performed the parasitological techniques. SM carried out the dissection, morphological and taxonomical identification, and figure illustration. GM performed sample collection, preservation, and descriptive issues. DO, GM, and SMC conceived and designed the study and reviewed the final manuscript. All authors read and approved the final manuscript.

Data availability All data generated or analyzed during this study are included in this published article. The original datasets are available upon request to the corresponding author.

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Declarations

Ethics approval This sample was captured accidentally during an authorized rodent pest control program that was done under the Ahvaz City municipality regulations in the late 1990s. Due to the delayed trap collection, we confronted the dead carcass.

Consent to participate Not applicable.

Consent for publication Not applicable.

Conflict of interest The authors declare no competing interests.

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