

Protozoan and Nematode Parasites of the American Coakroach *Periplaneta americana* (L.) from Bulgaria

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Abstract: Protozoan and nematode infections of *Periplaneta americana* in Bulgaria were investigated. Five protozoan and 2 nematode parasites of *P. americana* were recorded: *Endamoeba blattae*, *Amoeba* sp., *Lophomonas striata*, *Nyctotherus ovalis*, *Balantidium blattarum*, *Hammerschmidtella diesingi* and *Leidynema appendiculatum*. Parasite prevalence varied from 1,6% for *Amoeba* sp. to 32,8% for nematodes. Biometrical data for the established nematode species, compared with data reported by other authors are also presented.

Key words: *Periplaneta americana*, protozoan parasites, parasitic nematodes, prevalence, Bulgaria

Introduction

The American cockroach (*Periplaneta americana* L.), is worldwide synantropic species, whose populations live in warm moist habitats. It was first reported in Bulgaria by DRENSKY as an alien invasive species (1938). The cockroach is found in restaurants, enterprises for food industry, grocery stores and other places, but it can also be observed outside residential areas, like dumps, under firewood, mines and in unheated homes. During the summer months it can be seen in open areas – in yards or in the streets. In Bulgaria it occurs most often in substations of residential blocks and sewage systems.

American cockroaches can become a public health problem due to their association with human waste and diseases, and their ability to move from sewers into homes and commercial establishments. Their presence in these habitats is of epidemiological significance (BELL, ADIYODI 1981).

Protozoan and nematode parasites of *P. americana* were investigated by many parasitologists from

the US, Europe and Asia (GHOSH 1922a,b, BASIR 1940, HOYTE 1961, DESPORTES 1966, BRISCOE 1971, ALI *et al.* 1984, ANUAR 1987, ZERVOS 1988a,b, etc.). At least 30 species of protozoans (genera: *Balantidium*, *Endamoeba*, *Endolimax*, *Entamoeba*, *Iodamoeba*, *Lophomonas*, *Nyctotherus*, *Isotricha*, *Polymastix*, *Gregarina* and others) and more than 10 species of helminths (genera: *Binema*, *Gordius*, *Neoplectana*, *Thelastoma*, *Hammerschmidtella*, *Leidynema*, *Prostrellus*) have been isolated from field collected American cockroaches as a primary host. Seven protozoan genera (genus *Chilomastix*, *Entamoeba*, *Giardia*, *Nosema*, *Tetrahymena*, *Trichomonas*) caused infection in *P. americana* in experimental conditions (BELL, ADIYODI 1981). According to MAKETON *et al.* (2010) some entomopathogenic nematodes can be used for the biological control of *P. americana*. So far, in Bulgaria no studies of protozoan and nematode parasites of *P. americana* have been conducted.

The aim of the present study was to investigate the protozoan and nematode infections of *P. americana* in Bulgaria and compare their prevalence with other investigated cockroaches from the country.

Material and Methods

The subjects of our investigations were living specimens of *P. americana*, collected from the city of Sofia, where the species was detected. In total, 61 specimens of cockroaches were dissected in 2010 and 2012. Fresh preparations of gonads, Malpighian tubules, body fat, and the entire gut of the host were observed under light microscopy at magnification 100x and 400x for the presence of parasites. Slides were contrasted and stained with Lugol solution and Acetocarmine.

Observed nematodes, isolated from the intestine of the insects were heat killed (65° C), fixed in TAF (mixture of trietanolamine, formalin and water), processed in glycerin (SEINHORST 1959) and studied on permanent mounts. Biometrical measurements of the found nematode larvae as well as of adult males and females were conducted. Pictures of the observed parasites were made with a digital Camera Olympus. The permanent preparations were deposited at the Institute of Biodiversity and Ecosystem Research, BAS.

Results and Discussion

As a result of our study the following 5 protozoan and 2 nematode parasites of *P. americana* were recorded:

Amoebozoa

Endamoeba blattae (Butschli) was observed in the gut lumen of 3 of the investigated individuals (4.9%). The species was reported by many authors in *Blata orientalis*, *Blatella germanica* and *P. americana* (Hoyte 1961, Lipa 1975).

Amoeba sp. (Fig. 1 A). In one adult insect (1.6%), collected in 2010, several big mononuclear active amoebae were observed, whose dimensions varied from 80-150 µm to 80-120 µm. No cysts were found in the preparation. We suppose it's a case of free-living *Amoeba* sp. developing as a commensal organism in the colon of the omnivorous *P. americana*.

Chromalveolata

Lophomonas striata STEIN (Fig. 1 B) was found in 3 of the individuals studied (4.9%). It was recorded also in *B. orientalis*, *B. germanica* and *P. americana* (SPRAGUE 1941, HOYTE 1961, TSAI, CAHIL

1970). In Bulgaria, LIPA *et al.* (1991) reported it in *Blatta orientalis*.

Nyctotherus ovalis LEYDI (= *Nicthotherus periplanetae* Lalpatu) (Fig. 1 C) was observed in the gut lumen of 22 of the studied individuals (36.1%). It has been reported in *B. germanica* and *P. americana* by HOYTE (1961), TSAI, CAHIL (1970). LIPA *et al.* (1991) found it in *B. orientalis* in Bulgaria.

Balantidium blattarum GHOSH was detected in 9 of the investigated individuals (14.75%).

Nematoda, Oxyurida

Hammerschmidtia diesingi (HAMMERSCHMIDT, 1838) CHITWOOD, 1932 (Fig. 1 D, Fig. 2)

Synonyms – *Oxyuris diesingi* HAMMERSCHMIDT, 1838; *Oxyuris blattae orientalis* HAMMERSCHMIDT, 1847; *Streptostomum gracile* LEIDY, 1850; *Anguillula macrura* DISING, 1851; *Aorurus diesingi* (HAMMERSCHMIDT, 1838) TRAVASSOS, 1929.

Measurements – Table 1.

Description: Females (n = 5): L = 2.068 - 3.164 mm; a = 11-12.3 (11.7 ± 0.48); b = 6.4-10.4 (8.7 ± 1.47); c = 2.9-3.2 (2.9 ± 0.13); V = 21.3-25.6 (22.48 ± 1.79)%. Body spindle-shaped. Narrow lateral alae present. Oesophagust consisting from cylindrical procorpus, large ovoid metacarpus (pseudobulb), distinct isthmus which passes into valvate bulb. The lumen of procorpus and the first third of the metacarpus is strong sclerotized. Nerve ring situated on posterior end of procorpus. Excretory pore located at 434-485 µm from the anterior end. Vulva is a transverse slit in anterior third of the body, posterior to base of the oesophagus. A long vagina and common uterus posteriorly directed. Gonads didelphic, prodelphic. Eggs elongated, ellipsoidal. One pair of lateral pores at a distance of 20-50 µm after the anal opening. Tail long and filiform.

Males (n=2): L=0.824-0.836 mm; a=9.6-10.4; b = 5.2-5.4; c = 7.2-7.4. Body small, slightly curved ventrally at the posterior upon fixation. Cuticle finely striated up to the posterior region of corpus from head end. Esophagus consisting of procorpus, unclear metacarpus, isthmus and posterior valvular bulb. Testis and spicule single. Tail is clearly visible from the sharp contraction of the body, filiform shape. Caudal papillae consists of four pairs, one pair subventral pre-anal, one pair adanal, one pair subventral just posterior to anus and one pair at the base of the caudal appendage.

Habitat: Intestine.

Remarks: Studies of the Bulgarian specimens are consistent with the description and scope of the

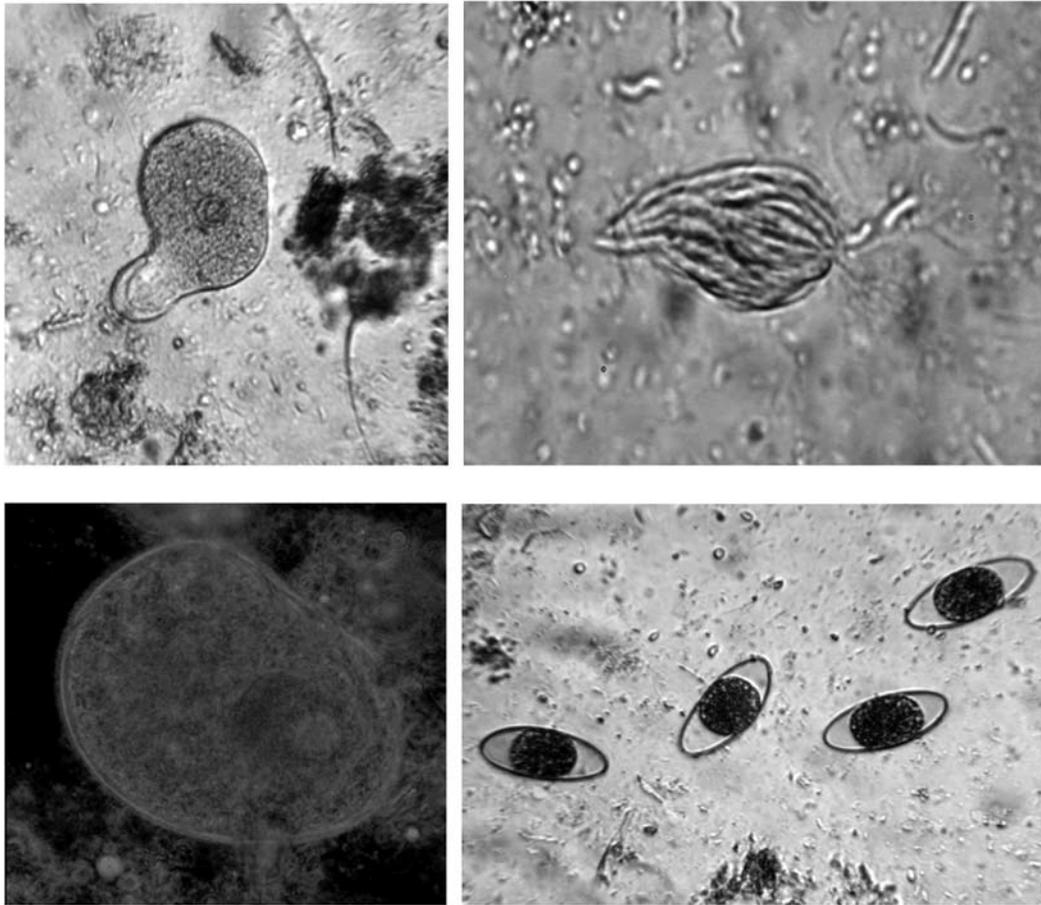


Fig. 1. A – *Amoeba* sp., B – *Lophomonas striata*, C – *Nyctotherus ovalis*, D – eggs of *Hammerschmidtella diesingi*

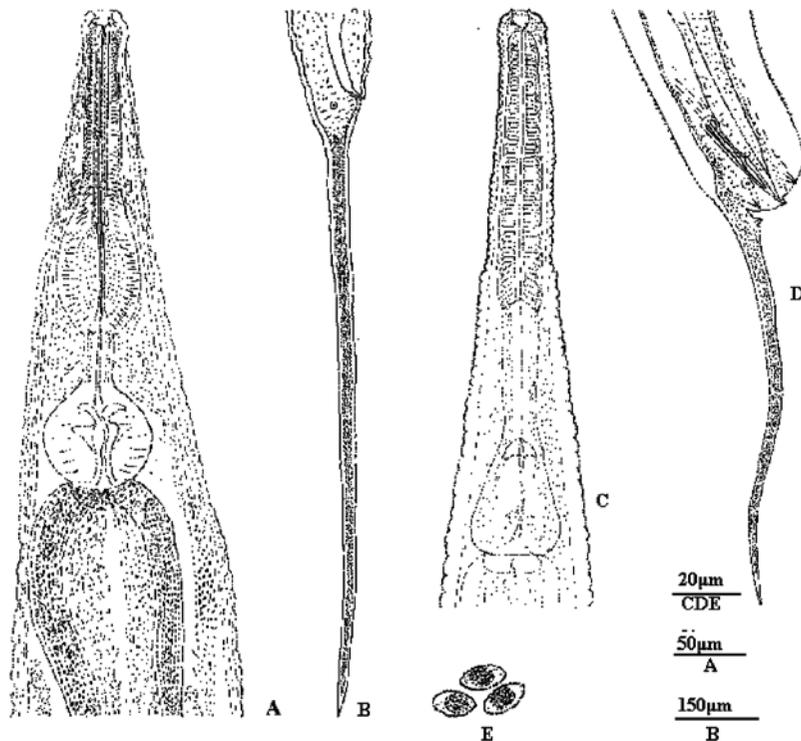


Fig. 2. *Hammerschmidtella diesingi* – Female: A – oesophageal region; B – tail; Male; C – oesophageal region; D – tail; E – eggs

Table 1. Measurements of *Hammerschmidtella diesingi*. All measurements are in mm unless otherwise mentioned

Autor	Nedelchev et al. (2012) Bulgaria	Chitwood (1932) USA	Leibersperger (1960) Germany	Shah (2007) India
Parasite	<i>H. diesingi</i>	<i>H. diesingi</i>	<i>H. diesingi</i>	<i>H. diesingi</i>
Host	<i>Periplaneta Americana</i>	<i>Periplaneta americana</i>	<i>Periplaneta americana</i>	<i>Periplaneta americana</i>
Female	n = 5		n = 39	n = 13
Length	2.068-3.164 (2.793±0.488)	No data	2.47-3.80	2.228-3.316
Width	0.168-0.284 (0.242±0.043)	No data	0.22 -0.48	0.166-0.284
Oesoph	0.303-0.352 (0.321±0.019)	No data	0.287-0.408	0.285-0.344
N.ring	0.090-0.124 (0.103±0.012)	No data	0.109-0.133	0.102-0.123
Excr. pore	0.434-0.485 (0.464±0.038)	No data	0.360-0.550	0.294-0.441
Vulva	0.530-0.672 (0.613±0.052)	No data	No data	0.441-0.834
Egg(in µm)	76-86(80±4) x 36-46(40±4)	No data	82-92 x 36-43	72.9-80.19x 29.16-34.02
Tail	0.686-1.090 (0.932±0.156)	No data	0.67 -1.07	0.756-0.991
Corpus	0.191-0.224 (0.207±0.015 x) 0.024-0.072	No data	0.181-0.237	0.182-0.223x 0.017-0.068
Isthmus	0.034-0.043 0.038±0.004 x 0.020-0.024 0.023±0.001	No data	0.034-0.077 0.014-0.024	0.034-0.051 0.019-0.024
End bulb	0.069-0.082 0.074±0.004 x 0.077-0.092 0.082±0.006	No data	0.072-0.094 0.068-0.104	0.060-0.080x 0.070-0.087
Pseudobulb	0.084-0.102 0.094±0.008 x 0.062-0.072 0.066±0.004	No data	0.072-0.104 0.062-0.082	0.072-0.099x 0.053-0.068
Male	n = 2	n = 2	n = 35	n = 7
Length	0.824-0.836	0.820-0.870	0.60-1.08	0.487-0.853
Width	0.080-0.86	0.052-0.064	0.040-0.110	0.034-0.082
Oesoph	0.152-0.156	0.126-0.190	0.125-0.169	0.116-0.145
N.ring	0.092-0.096	0.070-0.090	0.070-0.102	0.085-0.102
Excr. pore	0.208-0.222	0.200-0.330	0.220-0.250	0.136-0.179
Corpus	0.086-0.88 x 0.012-0.013	0.060-0.085 x 0.018-0.020	0.068-0.093 0.006-0.012	0.063-0.072x 0.007-0.010
Isthmus	0.036-0.039 0.010-0.11	No data	0.033-0.046 0.007-0.010	0.036-0.048x 0.007-0.009
End bulb	0.031-0.033 x 0.025-0.026	No data	0.024-0.030 0.019-0.026	0.019-0.029x 0.019-0.024
C. papillae	4 pairs	4 pairs	4 pairs	4 pairs
Spicule(in µm)	26-27.5	20-25	24-30	26.73-36.45
Tail	0.112-0.116	0.080-0.090	0.095-0.12	0.104-0.148
Gubernac.(in µm)		No data	13-19 ?	

measurements given by earlier workers who presented their areas of distribution (Table 1). A cap-like characteristic structure of the tail tip of the female Indian population (SHAH 2007) in our specimens was not observed.

Distribution: North and South America, North India, China, Russia, Europe (Germany, Nördlingen) (LEIBERSPERGER 1960), Poland (GABRYELÓW K., E. LONG 1986), North-East India (SHAH 2007).

Leidynema appendiculatum (LEIDY 1850) CHITWOOD 1932 (Fig. 3)

Synonyms – *Thelastomum appendiculatum* Leidy, 1850; *Oxyuris blattae* Hammerschmidt, 1847, of GALEB, 1878; *Oxyuris blattae orientalis* Hammerschmidt, 1847, of BÜTSCHLI, 1878; *Oxyuris blattae orientalis* Hammerschmidt, 1847, of MAGALHÃES, 1900; *Leidynema blattae-orientalis* Hammerschmidt, 1847, of SCHWENK, 1929

Measurements – Table 2.

Description: Male (n = 1): L = 0.550 mm; a = 12.8; b = 3.3; c = 39.9; tail = 13.8 µm; Body curved at the posterior end upon fixation. Cuticle strii 4.6-6 µm long in the area of the corpus, on the rest of the body 2-2.6 µm. Oesophagus 170 µm long by 10.5 µm wide, a short isthmus 21 µm long, 10.5 µm wide. Intestine simple, without diverticula and loop. Testis single, reflexed at the tip. Spicule 30 µm long. Tail short, rounded and offset addition with cone, provided with one pair of large subventral preanal papillae. Postanal four smaller pairs of papillae: three of

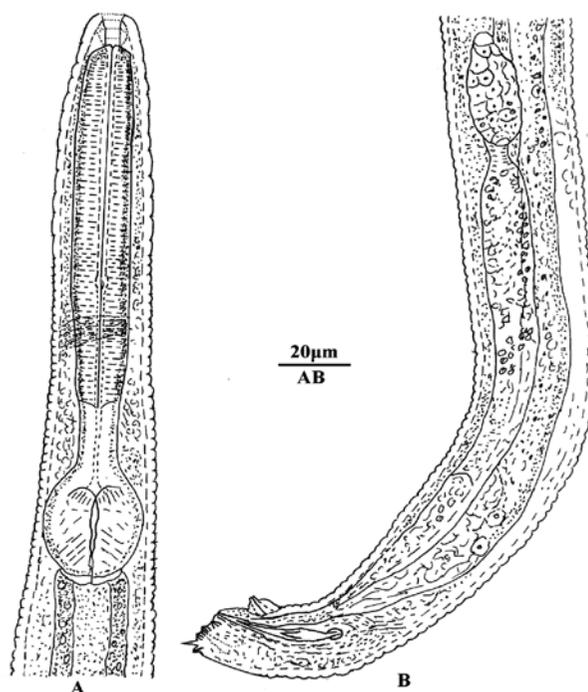


Fig 3. *Leidynema appendiculatum* – Male; A – oesophageal region; B – posterior region of body

them are nearly median, second and third pair almost merges into a double papilla. The fourth pair of dorso-laterally situated on slightly extended lobes.

Habitat: Intestine.

Distribution: North and South America, Europe (Germany), China, Russia (LEIBERSPERGER 1960), Poland (GABRYELÓW K., E. LONG 1986), North-East India (SHAH 2007).

Table 2. Measurements of *Leidynema appendiculatum* – male. All measurements are in mm unless otherwise mentioned.

Autor	Nedelchev <i>et al.</i> (2012) Bulgaria	Chitwood (1932) USA	Leibersperger (1960) Germany	Shah (2007) India
Parasite	<i>L. appendiculatum</i>	<i>L. appendiculatum</i>	<i>L. appendiculatum</i>	<i>L. appendiculatum</i>
Host	<i>Periplaneta americana</i>	<i>Periplaneta americana</i>	<i>Periplaneta americana</i>	<i>Periplaneta americana</i>
Male	n = 1	n = 2	n = 3	n = 8
L	0.55	0.840-0.880	0.52-1.28	0.579-1.119
tail	0.014	0.015-0.020	0.013-0.022	0.009 -0.012
Ex.pore	0.222	0.30-0.320	0.20-0.32	0.418
Nerve ring	0.090	0.12-0.155	0.07-0.155	0.108-0.127
Wide	0.043	0.050-0.060	0.06-0.12	0.058 -0.097
Corpus	0.117	0.140 -0.166	0.106	0.102 -0.123
Isthmus	0.021	No data	0.022	0.021-0.24
Bulb long wide	0.032 x 0.028	No data x 0.020 -0.030	0.040-0.046 x 0.036-0.042	0.034- 0.041 x 0.029-0.038
Spicule (in µm)	0.030	0.030	0.026-0.39	31.59-32.8

SHAN *et al.* (2007) investigated nematode fauna of *P. americana* from Manipur, India and established the following nematode species – *Thelastoma periplaneticola*, *L. appendiculatum*, *Protrellus shamimi* and *Hammerschmidtella diesingi*. These authors developed a determination key for these and other nematode genera parasitizing *Grylotalpa africana* and *Hydrophylus triangularis*.

AL-MAYALI, AL-YAQOUBI (2010) examined cockroaches and found 2 protozoans – *E. blatti* (33%), *N. ovalis* (65.3%) and 5 nematodes – *H. diesingi* (83.3%) *Thelastoma bulhoe* (15.4%), *Gordius robustus* (1.3%), *Enterobius vermicularis* eggs, (2%) *Ascaris lumbricoides* eggs (1.3%) from Iraq.

Recorded rates of infections with intestinal parasites in cockroaches from Bulgaria were comparable with the results published by AL-MAYALI, AL-YAQOUBI (2010), FOTEDAR *et al.* (1991) for the same host from Iraq. According to FOTEDAR *et al.* (1991) the prevalence rate of intestinal parasites of cockroaches was 99.4% in the cases where cockroaches were collected from hospitals and 94.2% in the cockroaches collected from residential areas. These val-

ues were higher than those reported by THYSSEN *et al.* (2004) who found 58.3% infections in *P. Americana*. GABRYELÓW, LONC, (1986) established that all 52 investigated specimens of *P. americana* from a laboratory stock of the Institute of Microbiology (Wroclaw, Poland) were infected with ciliates (probably *N. ovalis*) and 87% were infected with nematodes. In our study the infection rate caused by *N. ovalis* and nematodes was lower although the number of the investigated larvae was similar (61). The reason for this difference might be that all the individuals, which were studied by ABRYELÓW, LONC, (1986), originated from the same stock and could easily have gotten an infection via horizontal transmission.

Our parasitological investigations of *P. americana* show that this insect harbors several protozoan and nematode parasites and that this fact corroborates with the studies of other authors in Europe, USA and Asia.

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