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Special aspects of exogenic development of *Nematodirus spathiger* (Nematoda, Molineidae) at daily tempreture ranges

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Abstract

Ecological peculiarities in development of exogenous stages of *Nematodirus* nematodes include some important aspects of their survival and distribution at different abiotic factors. The aim of this work was laboratory study of some biological peculiarities of *Nematodirus spathiger* exogenous stages (Railliet, 1896) taken from cattle and grown at different temperatures (+20°C, 23°C, 26°C, 29°C and 32°C). The temperature was found significantly affects the period of exogenous development of *N. spathiger* eggs, both until L3 and on individual stages. Temperature also effects nematodes survival both eggs and hatched L3. At 32°C, the survival rates of eggs and hatched L3 were the highest, with the shortest period of development. At this temperature, L3 hatching was completed by day 10, and their survival was 94.3%. As temperature decreased, the time of L3 exogenous development prolonged, and survival rates were less: at 29°C – 12 days and 91.9%, at 26°C – 18 days and 86.7%, at 23°C – 22 days and 80.0%, at 20°C – 30 days and 56.7%, respectively. During the development of *N. spathiger* eggs, their length and shell thickness decreased by 10.6 and 22.6%, respectively, and their width increased by 10.5%. In addition, changes in morphometric parameters during L3 maturation were observed, length and percent of filament to STE increased by 7.1 and 7.8%, and body width with and without sheath, and STE length decreased by 40.4%, 37.9%, and 8.8%, respectively.

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Introduction

Nematodirus nematodes are one of the main components of the widespread and practically significant trichostrongylid fauna in digestive tract of domestic and wild herbivores (1-3). Many of researchers study the fauna and distribution of these nematodes in sheep where *N. battus* Crofton & Thomas, 1951 (4-6), *N. oiratianus* Rajewskaja, 1929 (7), *N. abnormalis* May, 1920 (8), *N. spathiger* (Railliet, 1896) Railliet & Henry, 1909 (9,10), *N. filicollis* (Rudolphi, 1802) Ransom, 1907 (11) were identified and described. In contrast, only a few studies reported nematodiruses in cattle. Thus, in Canada, the nematodirosis infestation of cattle of different ages ranged from 0.7 to 40.0% (12,13). In the USA, *Nematodirus* spp. eggs were found coproscopically in 18% of examined calves (14). And in Afyonkarahisar province, Turkey, *Nematodirus* spp. was revealed in 16.49% of examined cattle (15).

One of the factors of the wide distribution of these nematodes is that their peculiarities of exogenous development significantly different from other trichostrongylids. The larvae (L1-L2-L3) of *Nematodirus* spp. develop inside eggs and hatch as invasive larvae L3 resistant to adverse environmental factors (16). Despite the lower fertility of *Nematodirus* species, as compared to other trichostrongylids, with only 20–30 eggs per day (17), their environmental resistance and survival is much higher because after exogenous development from eggs L3 hatch as larvae significantly resistant to abiotic factors (18).

Important, that development and survival of *Nematodirus* eggs and larvae requires certain temperatures. The relevant studies were made on parasites taken mainly from sheep. For hatching L3 from *N. filicollis* eggs, they need to be cooled below 10°C with maximum hatching near 4°C. The authors note that in the field L3 *N. filicollis* takes more than one year to hatch, and, probably, this may be an adaptive capacity allowed early infection of susceptible hosts in competition with *N. battus*, and their preservation in regions without proper eggs cooling (19). At the same time, L3 hatch from *N. battus* eggs all year round with autumn and spring peaks (20). Other researchers state that sensitization at low temperatures and subsequent incubation at higher temperatures are necessary for hatching of L3 *N. battus* similarly to *N. filicollis* (21,22).

Some reports told that L3 hatching from *N. spathiger* eggs from sheep need no preliminary cooling. Larvae emerge from eggs immediately after L3 formation (23). There was also reported about *N. spathiger* egg and L3 development in all seasons, although the period between laying eggs and hatching in colder months was longer than that in summer (24). More detailed studies of *N. spathiger* embryogenesis showed that eggs development is completely inhibited at 0°C. Inhibition of egg development at morula stage was also observed from 4 to 7°C and above 39°C with subsequent redevelopment at temperatures from 10 to 39°C. L3 hatching normally is from 21 to 36°C with the optimum 33°C giving the largest number of invasive larvae. The authors revealed that cooling of *N. spathiger* eggs to 0°C before culturing at 36°C has positive effect on subsequent development and survival of eggs and larvae with higher percentage of L3 hatching (25).

The aim of this work was laboratory examination at different temperatures of biological peculiarities of development on exogenous stages *Nematodirus spathiger* (Railliet, 1896) Railliet and Henry, 1909 taken from cattle.

Materials and methods

Ethical approval

This study was conducted with the approval of Research Ethic Committee of the Poltava State Agrarian University, under protocol number /7/9/23.

Sample collection

Experimental studies were made during 2023–2025 in the Laboratory of Parasitology of the Poltava State Agrarian University (Ukraine). *N. spathiger* nematodes were collected in the course of complete helminthological dissection of cattle small intestines. Nematodes were identified to species by their morphometric parameters with keys (26,27). In laboratory, eggs were obtained from *N. spathiger* female gonads and washed into Petri dishes (not less than 70 specimens). Each egg culture (*N. spathiger* eggs *in* 0.9% NaCl solution) was cultivated at five different temperatures: 20°C, 23°C, 26°C, 29°C and 32°C until the formation and hatching of L3. Every 2 days, in experimental test cultures morphological changes in eggs, larvae, and their developmental stage were observed. The number and percentage of eggs on each developmental stage, number of L3 after hatching were counted, as well as mortality of eggs and L3. In experiment, *N. spathiger* eggs were cultivated in three replicates.

Microscopic examination and morphometric study

Morphometric parameters of *N. spathiger* eggs (length, width, shell thickness) and L3 (Figure 1) during their growth and development were counted using ToupView program version × 64, 4.10.17015.20200426 (Hangzhou ToupTek Photonics Co., Ltd, China). Microphotographs were made using SIGETA M3CMOS 14000 14.0 MP digital camera (China).

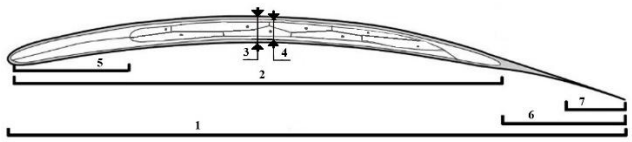

Figure 1: Metric parameters of L3 *Nematodirus spathiger* during cultivation: 1 – total length of sheath; 2 – length of larval body without sheath; 3 – maximum body width with sheath; 4 – maximum body width without sheath; 5 – length of larval esophagus; 6 – length of sheath tail extension (STE); 7 – length of filament.

Statistical analysis

Standard deviation (SD) and average values (M) were calculated. Significance of difference between average values in *N. spathiger* eggs studied and between L3 was established using one-way analysis of variance and F-test for 95% confidence level (28).

Results

During *N. spathiger* eggs cultivation regardless of temperature, the following developmental stages were identified according to morphological features: 1 blastomere formation (Figure 2a), 2 blastomeres formation (Figure 2b),

4 blastomeres formation (Figure 2c), 8 and more blastomeres formation (Figure 2d), formation of morula (Figure 2e), formed morula (Figure 2f), formation of L1 (Figure 2g), formed L1 (Figure 2h), formation of L2 (Figure 2i), formed L2 (Figure 2j), formed L3 inside egg (Figure 2k), L3 after hatching (Figure 2l), and invasive L3 with well-formed intestinal cells (Figure 2m).

The duration of *N. spathiger* eggs development and L3 hatching and their survival at different temperatures varied significantly. At 20°C, the largest number of hatched L3 was on day 30 when 56.7% of viable L3 were seen (Table 1, Figure 3).

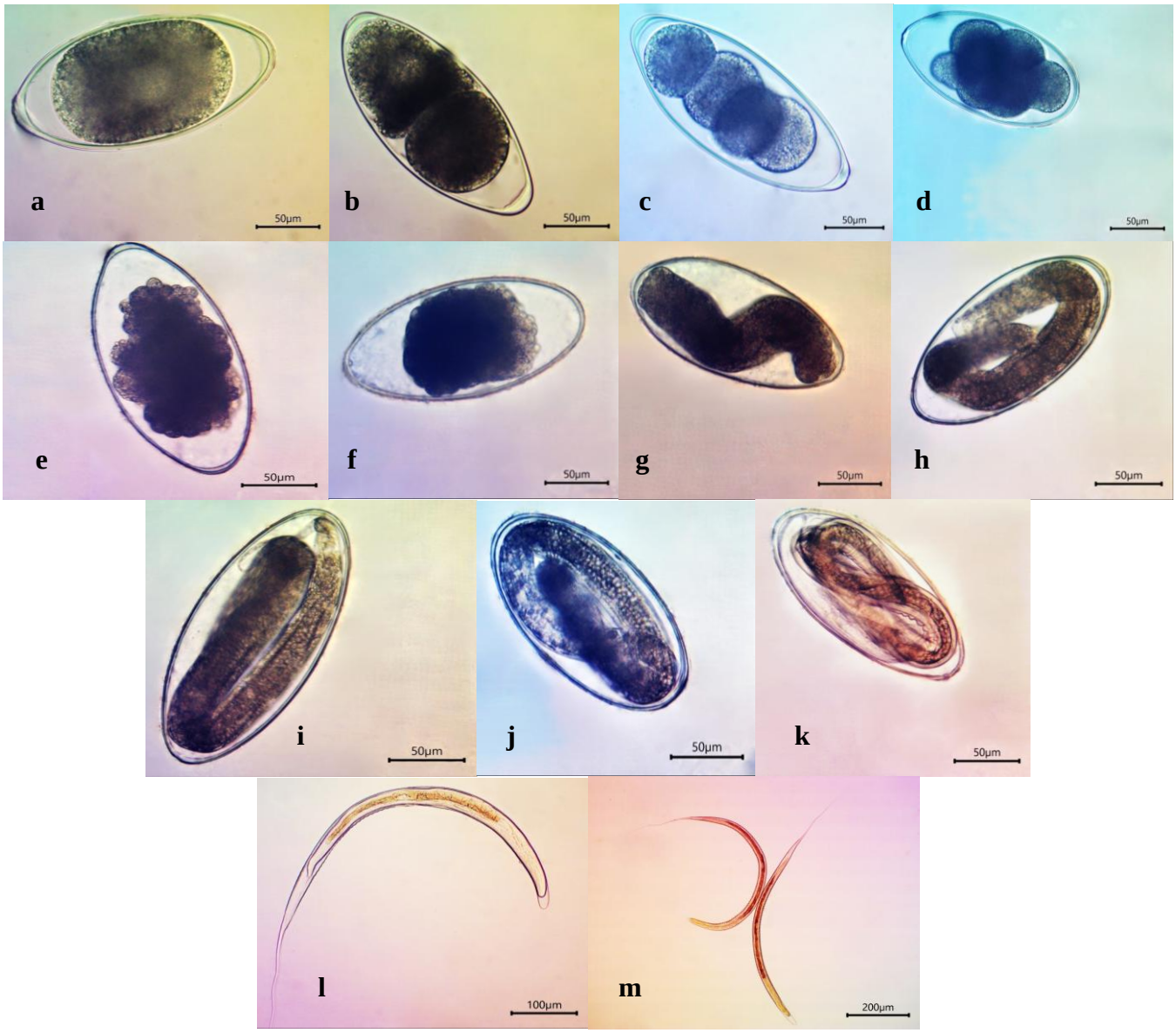

Figure 2: Exogenous stages of *Nematodirus spathiger* development: 1 blastomere formation (a); 2 blastomeres formation (b); 4 blastomeres formation (c); 8 and more blastomeres formation (d); morula formation (e); formed morula (f), L1 formation (g); formed L1 (h); L2 formation (i); formed L2 (j); formed L3 inside egg (k); L3 after hatching (l); invasive L3 with formed intestinal cells (m).

Table 1: Developmental points of *Nematodirus spathiger* eggs cultivated at 20°C (n=70, M ± SD)

Day of study	Stage of development, individuals													
	1 blastomere	2 blastomeres	4 blastomeres	8 and more blastomeres	morula formation	formed morula	formed L1	formed L1	formation of L2	formed L2	formed L3 in egg	L3 after hatching	Death of eggs, ind.	Death of L3, ind.
0	6.0 ± 3.0	27.7 ± 14.2	23.7 ± 11.4	12.7 ± 8.3	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
2	0.0 ± 0.0	9.0 ± 4.0	11.3 ± 4.5	16.7 ± 2.1	26.7 ± 5.0	6.3 ± 3.1	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	9.3 ± 4.0	32.7 ± 2.5	20.7 ± 6.0	7.3 ± 2.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
6	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	15.7 ± 2.1	11.0 ± 3.6	11.3 ± 2.1	8.7 ± 1.5	5.3 ± 3.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	18.0 ± 2.6	0.0 ± 0.0
8	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	3.7 ± 2.5	9.0 ± 2.6	14.0 ± 4.0	12.0 ± 2.6	10.0 ± 5.6	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	21.3 ± 1.5	0.0 ± 0.0
10	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	7.0 ± 2.6	19.0 ± 2.0	18.0 ± 2.0	4.7 ± 2.5	0.0 ± 0.0	0.0 ± 0.0	21.3 ± 1.5	0.0 ± 0.0
12	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	2.0 ± 1.0	12.3 ± 2.5	20.7 ± 3.5	13.7 ± 1.5	0.0 ± 0.0	0.0 ± 0.0	21.3 ± 1.5	0.0 ± 0.0
14	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	7.7 ± 2.1	14.3 ± 2.1	20.7 ± 1.5	6.0 ± 3.0	0.0 ± 0.0	21.3 ± 1.5	0.0 ± 0.0
16	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	4.3 ± 1.2	7.0 ± 2.0	19.0 ± 3.0	18.3 ± 2.1	0.0 ± 0.0	21.3 ± 1.5	0.0 ± 0.0
18	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	2.0 ± 1.0	6.0 ± 1.0	16.3 ± 2.1	24.3 ± 1.5	0.0 ± 0.0	21.3 ± 1.5	0.0 ± 0.0
20	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	4.0 ± 1.0	10.0 ± 1.0	34.7 ± 1.2	0.0 ± 0.0	21.3 ± 1.5	0.0 ± 0.0
22	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	1.3 ± 0.6	8.0 ± 1.0	29.0 ± 3.0	4.7 ± 3.1	21.3 ± 1.5	5.7 ± 3.1
24	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	5.0 ± 1.0	22.0 ± 1.0	13.3 ± 3.5	21.3 ± 1.5	8.3 ± 1.5
26	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	2.3 ± 1.5	10.0 ± 2.0	27.3 ± 1.2	21.3 ± 1.5	9.0 ± 1.0
28	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	2.3 ± 1.5	37.3 ± 2.1	21.3 ± 1.5	9.0 ± 1.0
30	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	39.7 ± 2.5	21.3 ± 1.5	9.0 ± 1.0

Eggs with 1 blastomere were seen before cultivation only in 8.6%. Stage of 2 blastomeres was seen up to day 2 in quantity from 39.5 to 12.9%. Stages of 4 blastomeres and 8 and more blastomeres were 16.2–33.8% and 13.3–18.1%, respectively, during 2 and 4 days of cultivation. Morula formation and formed morula were 38.1–5.2% and 9.0–15.7%, respectively, during 2–6 and 2–8 days. L1 formation and formed L1 were 2.9–16.2% and 2.9–27.1%, respectively, during 4–12 and 6–18 days of cultivation. L2 formation and formed L2 were 1.9–29.5% and 3.3–29.5%, respectively, during 6–22 and 10–26 days of cultivation. L3 formation inside eggs was observed from day 14 in 8.6% with the maximum on day 20 (49.5%), however on day 28 they were only in 3.3%. L3 hatching began from day 22 (6.7%) and their number gradually increased until day 30. *N. spathiger* eggs mortality was 30.5%, post-hatching L3 mortality 12.9%.

At 23°C, the number of hatched and viable L3 increased to 80.0%, their hatching time reduced to 22 days (Table 2, Figure 4).

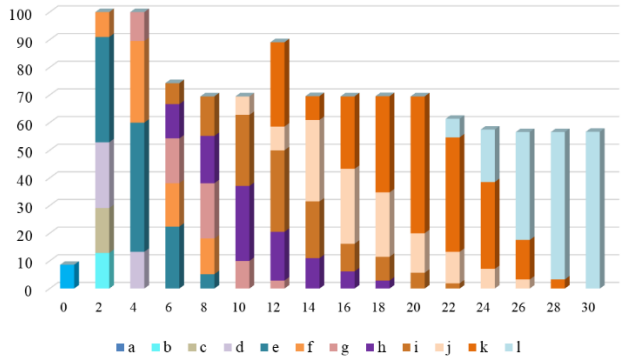

Figure 3: The ratio of *Nematodirus spathiger* exogenous stages during cultivation at 20°C (%): notes see in Figure 2.

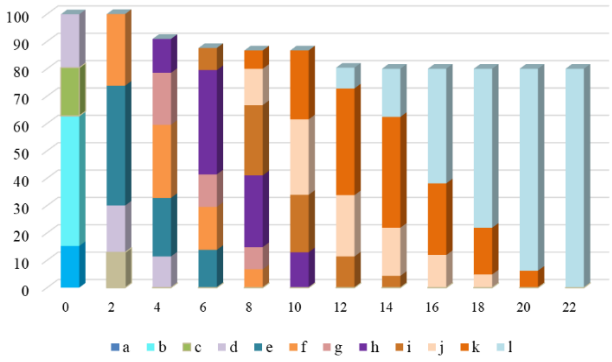

Figure 4: The ratio of *Nematodirus spathiger* exogenous stages cultivated at 23°C (%): notes see in Figure 2.

Table 2: Developmental points of *Nematodirus spathiger* eggs cultivated at 23°C (n=70, M ± SD)

Day of study	Stage of development, individuals												Death of eggs, ind.	Death of L3, ind.
	1 blastomere	2 blastomeres	4 blastomeres	8 and more blastomeres	morula formation	formed morula	formed L1	formed L1	formation of L2	formed L2	formed L3 in egg	L3 after hatching		
0	10.7 ± 3.5	33.3 ± 14.0	12.3 ± 7.4	13.7 ± 4.7	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
2	0.0 ± 0.0	0.0 ± 0.0	9.0 ± 4.0	12.0 ± 1.0	30.7 ± 2.1	18.3 ± 2.1	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	8.0 ± 3.6	15.0 ± 3.6	18.7 ± 4.7	13.3 ± 2.5	8.7 ± 3.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	6.3 ± 1.5	0.0 ± 0.0
6	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	9.7 ± 3.5	11.0 ± 2.0	8.3 ± 1.5	26.7 ± 6.1	5.7 ± 3.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	8.7 ± 1.5	0.0 ± 0.0
8	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	4.7 ± 2.5	5.7 ± 1.5	18.3 ± 1.2	18.0 ± 3.6	9.3 ± 1.5	4.7 ± 3.1	0.0 ± 0.0	9.3 ± 2.1	0.0 ± 0.0
10	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	19.0 ± 2.0	14.7 ± 1.5	19.3 ± 1.5	17.7 ± 3.1	0.0 ± 0.0	9.3 ± 2.1	0.0 ± 0.0
12	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	9.0 ± 4.0	8.0 ± 2.0	15.7 ± 1.5	27.3 ± 2.1	5.3 ± 1.5	9.3 ± 2.1	4.3 ± 1.5
14	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	3.0 ± 1.0	12.3 ± 1.5	28.3 ± 4.2	12.3 ± 2.5	9.3 ± 2.1	4.7 ± 1.2
16	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	8.3 ± 1.5	18.3 ± 2.5	29.3 ± 1.5	9.3 ± 2.1	4.7 ± 1.2
18	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	3.3 ± 1.5	12.0 ± 2.0	40.7 ± 2.1	9.3 ± 2.1	4.7 ± 1.2
20	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	4.3 ± 1.5	51.7 ± 1.2	9.3 ± 2.1	4.7 ± 1.2
22	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	56.0 ± 2.6	9.3 ± 2.1	4.7 ± 1.2

Stages of 1 and 2 blastomeres were seen before cultivation only, at the starting point their number was 15.2 and 47.6%, respectively. Stages of 4 blastomeres and 8 and more blastomeres were 12.9–17.6% and 11.4–19.5%, respectively, during 2 and 3 days of cultivation. Morula formation and formed morula were 13.8–43.8% and 6.7–26.7%, respectively, during 2–6 and 2–8 days. L1 formation and formed L1 were 8.1–19.1% and 12.4–38.1%,

respectively, during 4–8 and 4–10 days of cultivation. L2 formation and formed L2 were 4.3–25.7% and 4.8–27.6%, respectively, during 6–14 and 8–18 days of cultivation. L3 formation inside eggs was observed from day 8 in 6.7% with the maximum on day 14 – 40.5%. On day 20, they were in 6.2% only. L3 hatching began from day 12 (7.6%) and their number gradually increased until day 22. *N. spathiger* egg mortality was 13.3%, post-hatching L3 mortality 6.7%.

At 26°C, the number of hatched and viable L3 increased up to 86.7%, their hatching time reduced to 18 days (Table 3, Figure 5).

Table 3: Developmental points of *Nematodirus spathiger* eggs cultivated at 26°C (n=70, M ± SD)

Day of study	Stage of development, individuals													
	1 blastomere	2 blastomeres	4 blastomeres	8 and more blastomeres	morula formation	formed morula	formed L1	formed L1	formation of L2	formed L2	formed L3 in egg	L3 after hatching	Death of eggs, ind.	Death of L3, ind.
0	7.0 ± 3.6	12.0 ± 8.9	24.0 ± 8.5	27.0 ± 12.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
2	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	7.3 ± 4.2	19.3 ± 2.5	33.3 ± 5.7	10.0 ± 3.6	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	18.3 ± 2.3	18.7 ± 2.5	28.7 ± 3.5	9.7 ± 2.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	4.7 ± 1.5	0.0 ± 0.0
6	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	9.0 ± 3.0	14.7 ± 3.5	25.3 ± 2.5	15.3 ± 1.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	5.7 ± 1.5	0.0 ± 0.0
8	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	7.3 ± 2.1	15.0 ± 2.0	22.0 ± 2.0	12.3 ± 1.5	7.7 ± 1.5	0.0 ± 0.0	5.7 ± 1.5	0.0 ± 0.0
10	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	12.3 ± 3.5	27.3 ± 3.5	15.7 ± 2.1	9.0 ± 2.0	5.7 ± 1.5	0.0 ± 0.0
12	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	5.0 ± 2.0	15.7 ± 2.5	23.3 ± 2.1	17.3 ± 2.5	5.7 ± 1.5	3.0 ± 1.0
14	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	6.3 ± 3.1	17.3 ± 2.1	37.0 ± 2.6	5.7 ± 1.5	3.7 ± 0.6
16	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	11.3 ± 2.5	49.3 ± 2.1	5.7 ± 1.5	5.7 ± 1.5
18	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	60.7 ± 1.5	5.7 ± 1.5	5.7 ± 1.5

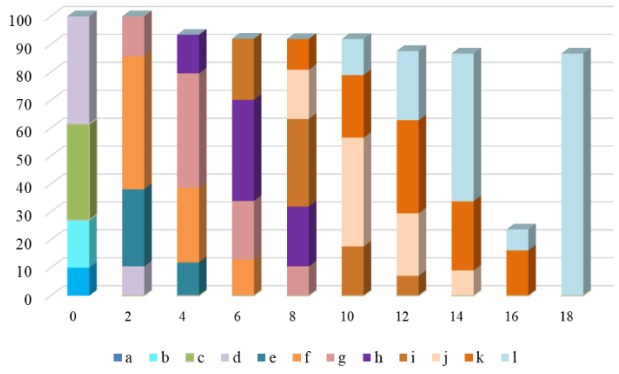

Figure 5: The ratio of *Nematodirus spathiger* exogenous stages cultivated at 26°C (%): notes see in Figure 2.

Stages of 1, 2 and 4 blastomeres were seen before cultivation only, at the starting point their number was 10.0%, 17.1% and 34.3%, respectively. Stage of 8 and more blastomeres was 10.5–38.6% during the first 2 days of cultivation. Morula formation and formed morula were 11.9–27.6% and 12.9–47.6%, respectively, during 2–6 days of cultivation. L1 formation and formed L1 were 10.5–41.0% and 13.8–36.2%, respectively, during 2–8 and 4–8 days of cultivation. L2 formation and formed L2 were 7.1–31.4% and 9.0–39.0%, respectively, during 6–12 and 8–14 days of cultivation. L3 formation inside eggs was observed from day 8 in 11.0% with the maximum on day 12 – 33.3%. On day 16, they were in 16.2%. L3 hatching began from day 10 (12.9%) and their number increased until day 18. *N. spathiger* eggs mortality was 8.1%, post-hatching L3 mortality 5.2%.

At 29°C, a greater number of *N. spathiger* embryonic and postembryonic stages survived with the shortened time of their development. The number of hatched L3 was 91.9% in 12 days (Table 4, Figure 6).

Table 4: Developmental points of *Nematodirus spathiger* eggs cultivated at 29°C (n=70, M ± SD)

Day of study	Stage of development, individuals													
	1 blastomere	2 blastomeres	4 blastomeres	8 and more blastomeres	formation of morula	formed morula	formation of L1	formed L1	formation of L2	formed L2	L3 formed in egg	L3 after hatching	Death of eggs, ind.	Death of L3, ind.
0	7.0 ± 3.6	12.0 ± 8.9	24.0 ± 8.5	27.0 ± 12.5	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
2	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	7.3 ± 4.2	19.3 ± 2.5	33.3 ± 5.7	10.0 ± 3.6	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	3.3 ± 2.3	18.7 ± 3.1	21.3 ± 1.5	24.0 ± 1.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	2.7 ± 1.2	0.0 ± 0.0
6	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	7.3 ± 3.5	17.7 ± 1.5	28.7 ± 3.2	12.3 ± 1.5	0.0 ± 0.0	4.0 ± 1.0	0.0 ± 0.0
8	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	5.7 ± 3.5	49.7 ± 8.7	9.0 ± 4.6	4.0 ± 1.0	1.7 ± 0.6
10	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	21.0 ± 2.0	43.3 ± 2.1	4.0 ± 1.0	1.7 ± 0.6
12	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	64.3 ± 1.5	4.0 ± 1.0	1.7 ± 0.6

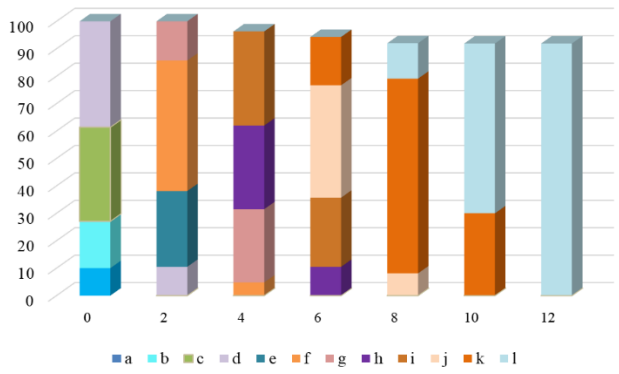

Figure 6: The ratio of *Nematodirus spathiger* exogenous stages cultivated at 29°C (%): notes see in Figure 2.

Stages 1, 2 and 4 blastomeres were seen before cultivation, their number was 10.0%, 17.1% and 34.3%, respectively. Stage of 8 and more blastomeres was 10.5–38.6% during the first 2 days of cultivation. The stages of morula formation and formed morula were 27.6% and 4.8–47.6%, respectively, during 2 and 2–4 days. L1 formation and formed L1 were 14.3–26.7% and 10.5–30.5%, respectively, during 2–4 and 4–6 days of cultivation. L2 formation and formed L2 were 25.2–34.3% and 8.1–41.0%, respectively, during 4–6 and 6–8 days. L3 formation inside eggs was observed since day 6 when their number was 17.6%. The maximum number was reached on day 8 – 71.0%. On day 10, their number decreased till 30.0%. L3 hatching began on day 8 (12.9%) and their number increased during the cultivation until day 12. *N. spathiger* eggs mortality was 5.7%, L3 post-hatching mortality 2.4%.

At 32°C, the largest number of *N. spathiger* hatched L3 was 94.3% in the shortest period of time – 10 days (Table 5, Figure 7).

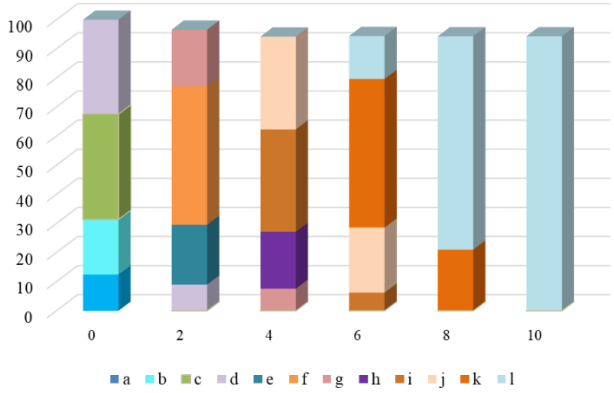

Figure 7: The ratio of *Nematodirus spathiger* exogenous stages cultivated at 32°C (%): notes see in Figure 2.

Table 5: Developmental points of *Nematodirus spathiger* eggs cultivated at 32°C (n=70, M ± SD)

Day of study	Stage of development, individuals												
	1 blastomere	2 blastomeres	4 blastomeres	8 and more blastomeres	formation of morula	formed morula	formation of L1	formed L1	formation of L2	formed L2	L3 formed in egg	L3 after hatching	Death of eggs, ind.	Death of L3, ind.
0	8.7 ± 3.5	13.3 ± 3.1	25.3 ± 3.1	22.7 ± 4.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0
2	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	6.3 ± 4.0	14.3 ± 3.5	33.3 ± 10.3	13.7 ± 5.7	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	3.0 ± 1.0	0.0 ± 0.0
4	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	5.3 ± 3.1	13.7 ± 1.5	24.7 ± 4.6	22.3 ± 3.1	0.0 ± 0.0	0.0 ± 0.0	4.0 ± 1.0	0.0 ± 0.0
6	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	4.3 ± 2.1	15.7 ± 2.5	35.7 ± 2.5	10.3 ± 2.1	4.0 ± 1.0	0.0 ± 0.0
8	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	14.7 ± 2.5	51.3 ± 1.5	4.0 ± 1.0	0.0 ± 0.0
10	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	66.0 ± 1.0	4.0 ± 1.0	0.0 ± 0.0

Stages of 1, 2 and 4 blastomeres were also seen before cultivation only with their number 12.4%, 19.0% and 36.2%, respectively. Stage of 8 and more blastomeres was 9.0–32.4% during the first 2 days of cultivation. The stages of morula formation and formed morula were 47.6% and 7.6–19.5%, respectively, during 2 and 2–4 days. L1 formation and formed L1 were 7.6–19.5% and 19.5%, respectively, during 2–4 and 4 days. L2 formation and formed L2 were 6.2–35.2% and 22.4–31.9%, respectively, during 4–6 days of cultivation. L3 formation inside eggs was observed since day 6 when their number was maximal (51.0%). On day 8 their number decreased till 21.0%. L3 hatching began from day 6 (14.8%) and their number increased during the cultivation until day 10. *N. spathiger* eggs mortality was 5.7%. L3 post-hatching mortality was not revealed, all larvae remained viable.

The average metric parameters of immature *N. spathiger* eggs: length – 209.2 μm, width – 90.2 μm, shell thickness – 3.1 μm (Table 6).

Table 6: Metric parameters of *Nematodirus spathiger* eggs (own and literature data)

Author	Parameters, μm					
	Egg length		Egg width		Eggshell thickness	
	Mean	Range	Mean	Range	Mean	Range
Present specimens, n=50	209.2	176.3 – 244.9	90.2	82.7 – 95.9	3.1	2.5 – 3.6
Railliet et al., 1912 (29)	–	200 – 260	–	100 – 110	–	–
May, 1920 (30)	–	150 – 220	–	80 – 110	–	–
Shore, 1939 (31)	202.0	181 – 230	98.3	91 – 107	–	3.3 – 3.8
Tetley, 1935 (32)	–	180 – 210	–	90 – 105	–	–
Tetley, 1941 (33)	195.0	179 – 210	97.0	88 – 110	–	–
Skrjabin et al., 1954 (26)	–	221 – 238	–	119 – 136	–	–
Kates & Turner, 1955 (34)	200.0	–	98.0	–	–	–
Thomas, 1957 (35)	–	183 – 214	–	87 – 99	–	–
Soulsby, 1968 (36)	–	175 – 260	–	106 – 110	–	–
Viljeon, 1972 (25)	218.0	173 – 238	103.0	97 – 119	–	–
Lichtenfels & Pillit, 1983 (37)	–	172 – 217	–	95 – 114	–	–
Ivashkin et al., 1981 (27)	–	221 – 238	–	119 – 136	–	–
Melnychuk et al., 2021 (9)	202.2	187.3 – 218.8	112.0	103.9 – 116.8	2.38	2.14 – 2.66

Note: "–" – parameters were not defined.

In the literature, there are some differences in metric parameters of *N. spathiger* eggs: the average egg length ranged from 195 to 218 μm, width - from 97 to 112.0 μm. The minimum and maximum values of shell thickness were from 2.14 to 3.6 μm.

We also found that during cultivation the size of nematodirus eggs changes ($P < 0.001$). As compared to similar data for egg cleavage and blastomeres formation, shell length and thickness of eggs with L3 inside appeared to be 10.6% less (Figure 8a) and 22.6% less (Figure 8c) while eggs width was 10.5% more (Figure 8b).

Eight metric parameters of *N. spathiger* invasive L3 were established as average values: total length of sheath – 1028.8 μm; length of larval body without sheath – 722.7 μm; maximum body width with sheath – 24.9 μm; length of sheath tail extension – 19.8 μm; length of esophagus – 181.3 μm; length of STE– 312.3 μm; length of filament – 220.9 μm; percent of filament to STE – 70.8% (Table 7).

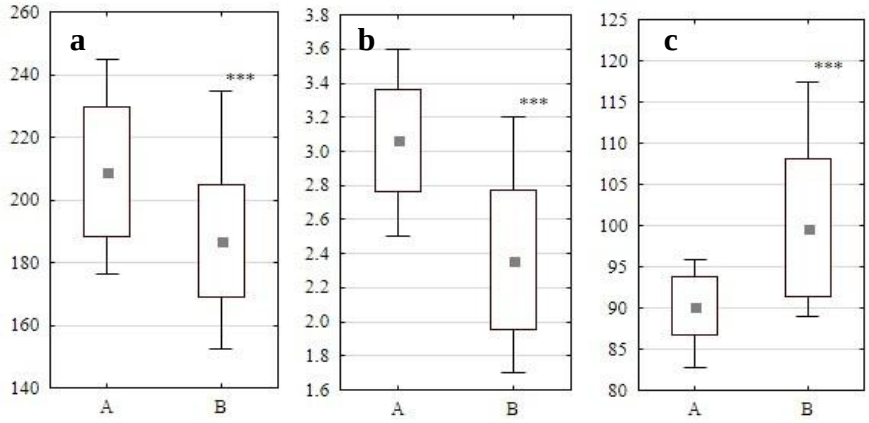

Figure 8: Metric parameters of *Nematodirus spathiger* eggs during cultivation at 32°C (μm): length (a); width (b), shell thickness (c); egg cleavage and blastomeres formation (A); L3 formation (B); *** – $P < 0.001$ – relative to data for egg cleavage and blastomeres formation (n=50).

Table 7: Metric parameters of L3 *Nematodirus spathiger* (own and published data)

Parameters, mkm (Mean, Range)	Present specimens, n=15	Monning 1931 (38)	Dikmans & Andrews, 1933) (39)	Thomas, 1957 (35)	Viljeon, 1972 (25)	Vanwyk et al., 2004; Van Wyk & Mayhew, 2013 (40,41)	Saidi et al., 2020 (3)
Total length of sheath	1028.8 931.9 – 1093.7	1158 1119 – 1117	1009 922 – 1118	1018 976 – 1130	1075 969 – 1282	–	1020
Length of larval body without sheath	722.7 632.5 – 845.1	–	–	151 723 – 810	–	–	–
Maximum body width with sheath	24.9 20.2 – 29.6	–	–	–	–	–	25
Maximum body width without sheath	19.8 17.6 – 22.9	–	–	–	–	–	–
Length of larval esophagus	181.3 146.4 – 219.3	235 227 – 235	195 160 – 225	204 192 – 224	188 103 – 235	–	–
Length of STE	312.3 242.2 – 362.7	340 325 – 340	328 315 – 350	332 310 – 390	333 270 – 416	270 267 – 309	–
Length of filament	220.9 189.8 – 259.7	–	–	–	–	–	–
% of filament to STE	70.8 66.0 – 78.8	–	–	–	–	60	–

Note: "–" – parameters were not defined.

In the literature available, there are from 2 to 6 parameters of L3 *N. spathiger*, including the average total length of sheath (1009-1158 μm), length of larval body without sheath – 151 μm, maximum body width with sheath – 25 μm, length of esophagus –188-235 μm, length of STE – 248-340 μm, percent of filament to the STE – 60%.

We also found that during maturation metric parameters of hatched L3 change. Particularly, invasive L3 length without sheath increases by 7.1% (722.7±54.8 μm, $P < 0.05$) (Figure 9b), maximum body width with sheath reduces by 37.9% (24.9±2.8 μm, $P < 0.001$) (Figure 9c) and maximum body width without sheath by 40.4% (19.8±1.6 μm, $P < 0.001$) (Figure 9d), length of STE decreases by 8.8% (312.3±27.5 μm, $P < 0.05$) (Figure 9f), percent of filament to STE increases by 7.8% (70.8±4.0%, $P < 0.001$) (Figure 9h).

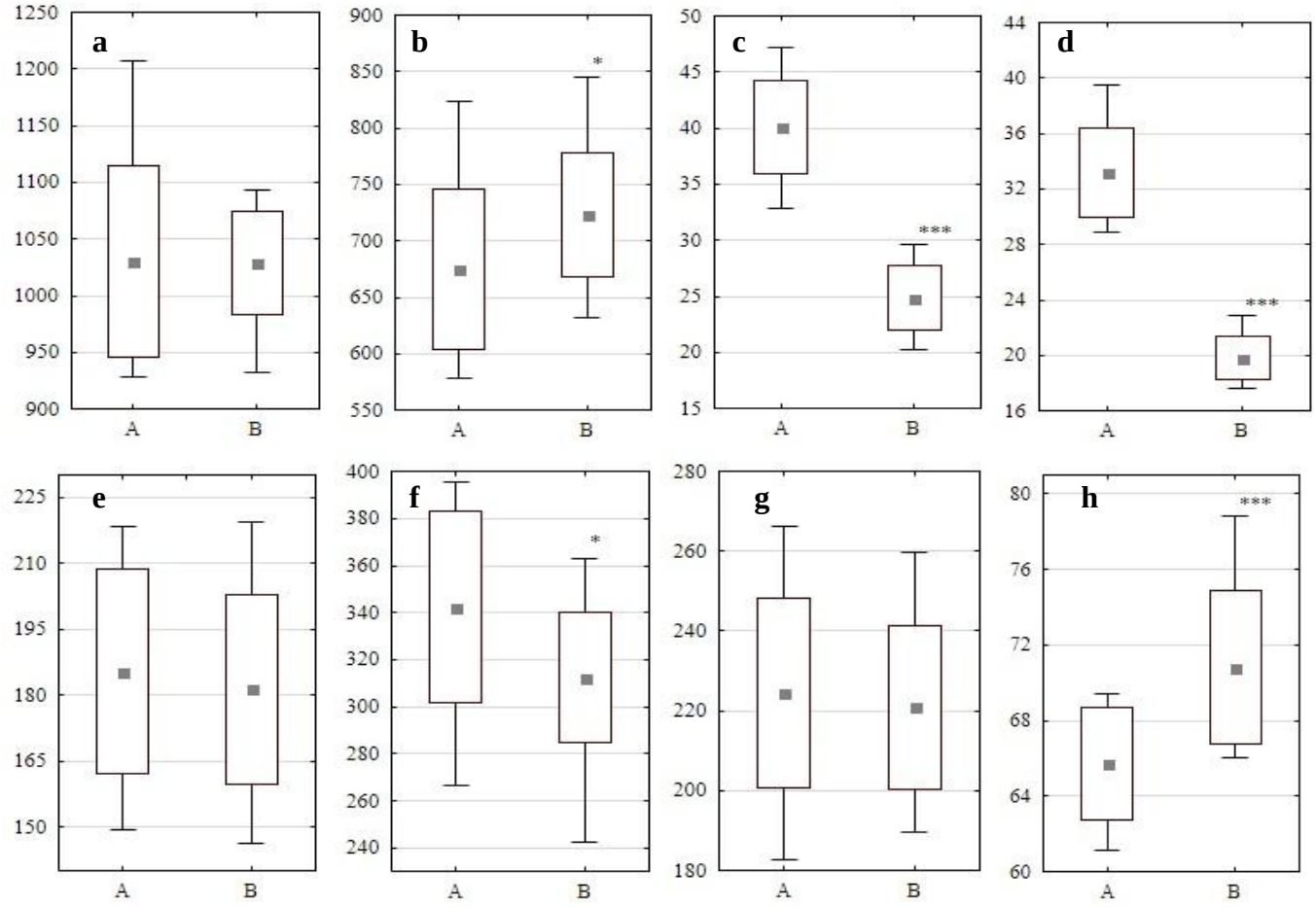

Figure 9: Metrics parameters of L3 *Nematodirus spathiger*: total length of sheath (a); length of larval body without sheath (b); maximum body width with sheath (c); maximum body width without sheath (d); length of larval esophagus (e); length of STE (f); percent of filament to STE (h); hatching stage (A); invasive stage (B); * – $P < 0.05$, *** – $P < 0.001$ – relative to data for hatching stage (n=15).

Discussion

Most studies demonstrated that nematodes of genus *Nematodirus* are widespread in ruminants with rather high infestation rate (2,42). One of the factors facilitating the survival of strongylids in digestive tract is the peculiarities of their biology especially on exogenous stages of their development. Moreover, unlike other strongylids, in *Nematodirus* nematodes larvae develop inside eggs and hatch into external environment as L3 (32,39). Therefore, for the first time in Ukraine, we study in laboratory the effect of temperature on period of development and survival of eggs in *N. spathiger* nematodes taken from cattle, as well as morphometric parameters of eggs and L3 during their growth and development.

Our results showed that during exogenous development of *N. spathiger*, a sequence of stages occurs. According to morphological characteristics we identified the following ones: 1, 2, 4, 8 and more blastomeres, formation of morula, morula, formation of L1, L1, formation of L2, L2, L3 inside

egg; L3 after hatching; invasive L3 with formed intestinal cells. Similar data were in other authors who determined such stages as: egg with 1, 4, 8 blastomeres, transition between blastomeres and morula, morula, tadpole stage (early first stage larvae), L1, transition between first and second stages, L2, L3 inside egg (25). There is a report where development of *Nematodirus* spp. consists of egg cells cleavage, egg cells begin to fuse, embryo entering the tadpole stage, L1 larva forming, L1 larva developing, L2 larva forming, L2 larva developing, the larva gradually developing into L3 larva, the larva hatching into mature L3 larva (7). At the same time, other authors note the following stages in the development of *Nematodirus helvetianus* May, 1920: 8 cells, 32 to 64 cells, morula, blastula and gastrula, tadpole, first-stage larva, first-stage larva in lethargus, second-stage larva, second-stage larva in lethargus, third-stage larva, hatching commences. Moreover, for the first time we found two stages of L3: L3 after hatching without formed intestinal cells and invasive L3 with formed intestinal cells.

As temperature increased, the period of egg development and L3 hatching in *N. spathiger* reduced, and their survival increases. The most optimal temperature was 32°C when development is rather quick: L3 hatch by day 10 at the highest number, 94.3%. As temperature decreased, development of *N. spathiger* exogenous stages becomes longer and the survival rate of both eggs and hatched L3 is lower. At temperature 29°C, L3 hatching finished by day 12 and their survival is 91.9%. At 26°C, 86.7% of L3 hatch in 18 days. At 23°C and 20°C, L3 hatch in 22 and 30 days, and their survival decreased to 80.0 and 56.7%, respectively.

Minor differences were found in works of researchers who observed the highest percentage of hatching L3 *N. spathiger* from sheep at temperatures from 27°C to 33°C. Moreover, at 33°C, the time to hatching of larvae was 8 days, so the authors concluded that this temperature is optimal for development and hatching of larvae, because in 21 days, 93% of viable larvae can hatch (25). Other authors cultivated *N. spathiger* eggs isolated from antelopes at 27°C and observed L3 hatching in 14 days (3). In *N. helvetianus* nematodes, at optimal temperature is 28°- 29°C, L3 can hatch after 8–10 days of cultivation (43). Such differences, in our opinion, may be due to specific developmental features depending on the species of *Nematodirus*, and host species of pathogen.

We compared our own data of the metric parameters of eggs and L3 with the data of other authors. We found that length of *N. spathiger* eggs at the blastomere cleavage stage ranged from 176.3 to 244.9 3.1 μm, width from 82.7 to 95.9 μm, and shell thickness from 2.5 to 3.6 μm. Other authors reported data where the egg length was from 150 to 260 μm, width from 80.0 to 136.0 μm, and shell thickness from 2.14 to 3.8 μm (Table 6). We think such discrepancies are due to measuring eggs at different stages of their development. This is confirmed by the results of our studies where in eggs at the stage of L3 formation the length and shell thickness are smaller by 10.6 and 22.6%, respectively ($P < 0.001$), and the width is larger by 10.5% ($P < 0.001$) as compared to similar parameters in eggs at the stage of cleavage and blastomere formation.

We determined 8 metric parameters of invasive L3 *N. spathiger* characterizing the total length of sheath, length of larval body without sheath, maximum body width with and without sheath, length of esophagus, length of STE – 312.3 μm, percent of filament length to STE. Other researchers determined from 2 to 6 parameters in L3 *N. spathiger* characterized the total length of sheath, length of larval body without sheath, maximum body width with sheath, the length of esophagus, the length of STE and percent of filament to STE (Table 7). Our data are a little different than the data of other authors that is also due to maturation of L3 after hatching. We established morphometric changes in L3 during their growth and development. Morphologically, invasive L3 contain well-formed intestinal cells, and metrically, their length of filament and its percent to STE are larger by 7.1% and 7.8%, respectively, and maximum body width with sheath and without sheath and also STE length are smaller by 37.9%, 40.4% and 8.8%, respectively, as compared to similar parameters in L3 immediately after hatching. The fluctuations in morphometric parameters of eggs and L3 *N. spathiger* identified can be considered while identification of these nematodes.

Conclusion

Exogenous development of *Nematodirus spathiger* nematodes in laboratory is characterized by the development of L1, L2 and L3 inside eggs, and the periods of development of those stages, hatching and viability of L3 depend on temperature regimen. While temperature increased from 20°C to 32°C, the time of egg development reduced, and the number of hatched L3 increases. At a temperature 32°C hatching begins on the 6th day and on the 8th day the largest number of viable L3 hatches – 94.3%. And at 20°C hatching begins on the 22nd day and on the 30th day the smallest number of viable L3 hatches – 56.7%. It was determined that the morphometric parameters of eggs and L3 of *N. spathiger* change during their growth and development. In eggs, the length (by 10.6%) and shell thickness (by 22.6%) gradually decrease, and the width (by 10.5%) increases. In invasive L3, the length without sheath (by 7.1%) and percent of filament to STE (by 7.8%) increase, body width with sheath reduces (by 37.9%), and maximum width without sheath (by 40.4%), and length of STE reduces (by 8.8%).

Acknowledgment

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Conflict of interest

There is no conflict of interest.

Referens

1. Hoberg EP, Lichtenfels JR, Rickard LG. Phylogeny for genera of Nematodirinae (*Nematoda*: Trichostrongylina). J Parasitol. 2005;91(2):382–89. DOI: 10.1645/GE-3408
2. Abramatov M, Kuchboev A, Ruziev B, Sobirov K. Diversity of gastrointestinal nematodes in domestic ruminants of Uzbekistan. Pak J Zool. 2022;54(5):2445–48. DOI: 10.17582/journal.pjz/20210629120602
3. Saidi A, Mimouni R, Hamadi F, Oubrou W. Larval morphological characteristics of *Camelostrongylus mentulatus* and *Nematodirus spathiger*. Ukr J Vet Agric Sci. 2020;3(2):7–11. DOI: 10.32718/ujvas3-2.02
4. Smith HJ, Hines JG. *Nematodirus battus* in Canadian sheep. Can Vet J. 1987;28(5):256. [available at]
5. Melville LA, Innocent G, Van Dijk J, Mitchell S, Bartley DJ. Nematode management practices and *Nematodirus battus* control strategies on UK sheep farms. Vet Rec. 2021;189(9):e775. DOI: 10.1002/vetr.775
6. Nikbin S, Almasi F, Alenizi D, Jenvey C, Sloan S, Preston S, Piedrafita D, Jonsson N, Stear M. Heritability of *Nematodirus battus* fecal egg counts. Parasitology. 2022;149(4):1–7. DOI: 10.1017/S0031182022000014
7. Liu Y, Wang P, Wang R, Li J, Zhai B, Luo X, Yang X. Epidemiological investigation and drug-resistant strain isolation of *Nematodirus oiratianus* in sheep in Inner Mongolia. Animals. 2022;13(1):30. DOI: 10.3390/ani13010030
8. Louw JP. *Nematodirus abnormalis* in sheep in the south-western Cape Province. Onderstepoort J Vet Res. 1989;56(2):141–42. [available at]
9. Melnychuk V, Yevstafieva V, Pishchalenko M, Reshetylo O, Antipov A. Morphological identification of *Nematodirus spathiger*. Regul Mech Biosyst. 2021;12(1):121–27. DOI: 10.15421/022119
10. Beltrame MO, Moviglia GS, De Tommaso D, Quintana S. Gastrointestinal parasites of domestic sheep from Patagonia: a paleoparasitological approach. Vet Parasitol Reg Stud Rep. 2023;44:100915. DOI: 10.1016/j.vprsr.2023.100915
11. Oliver AM, Leathwick DM, Pomroy WE. Prevalence of *Nematodirus spathiger* and *Nematodirus filicollis* in New Zealand sheep farms. N Z Vet J. 2014;62(5):286–89. DOI: 10.1080/00480169.2014.920700
12. Jelinski M, Gilleard J, Rocheleau L, Royan G, Waldner C. Epidemiology of gastrointestinal nematodes in grazing yearling beef cattle in Saskatchewan. Can Vet J. 2017;58(10):1044–50. [available at]
13. Wills FK, Waldner CL, Campbell JR, Pollock C, Uehlinger FD. Gastrointestinal nematode prevalence and fecal egg counts in beef cattle from western Canada. Can Vet J. 2020;61(6):605–12. [available at]
14. Stromberg BE, Gasbarre LC, Ballweber LR, Dargatz DA, Rodriguez JM, Kopral CA, Zarlenga DS. Prevalence of internal parasites in beef cows in the United States. Can J Vet Res. 2015;79(4):290–95. [available at]
15. Sevimli FK, Kozan E, Köse M, Eser M, Çiçek H. Gastrointestinal nematodes and seasonal distribution in cattle in Turkey. Turkiye Parazitol Derg. 2007;31(1):51–56. [available at]
16. Tetley JH. Ecological studies on *Nematodirus* species in sheep in New Zealand. J Helminthol. 1935;13(1):41–58. DOI: 10.1017/S0022149X0000331X
17. Brunsdon RV. Significance of *Nematodirus* in New Zealand. N Z Vet J. 1967;15(6):105–08. DOI: 10.1080/00480169.1967.33704
18. Parkin JT. Effect of moisture on development and hatching of *Nematodirus battus* eggs. Parasitology. 1976;73(3):343–54. DOI: 10.1017/S0031182000047028
19. Van Dijk J, Morgan ER. Hatching behaviour of *Nematodirus filicollis* in co-infection with *Nematodirus battus*. Parasitology. 2009;136(7):805–11. DOI: 10.1017/S003118200900609X
20. Gibson TE, Everett G. Ecology of free-living stages of *Nematodirus battus*. Res Vet Sci. 1981;31(3):323–27. [available at]
21. Thomas RJ, Stevens AJ. Development of pasture stages of *Nematodirus battus* and *Nematodirus filicollis*. Parasitology. 1960;50:31–49. DOI: 10.1017/S0031182000025178
22. Christie MG. Hatching of *Nematodirus battus* and remarks on *Nematodirus filicollis*. Parasitology. 1962;52(3-4):297–313. DOI: 10.1017/S0031182000027189
23. Oliver AM, Pomroy WE, Ganesh S, Leathwick DM. Chilling requirements for hatching of *Nematodirus filicollis*. Vet Parasitol. 2016;226:17–21. DOI: 10.1016/j.vetpar.2016.06.017
24. Gibson TE, Everett G. Ecology of the free-living stages of *Nematodirus spathiger*. Res Vet Sci. 1982;32(1):35–38. [available at]
25. Viljoen JH. Morphology of the free-living stages of *Nematodirus spathiger* with observations on development under laboratory conditions. J S Afr Vet Assoc. 1972;43(1):87–94. [available at]
26. Skrjabin KI, Shikhobalova NP, Shul'ts RS. Essentials of nematology: trichostrongylids of animals and man. Moscow, USSR: Academy of Sciences; 1954. 503–505 p. [available at]
27. Ivashkin VM, Mukhammadiev SA. Key to the helminthes of cattle. Moscow, USSR: Academy of Sciences; 1981. 177–178 p.
28. Petrie A, Watson P. Hypothesis tests: the F-test. In: Petrie A, Watson P editors. Statistics for veterinary and animal science. 3rd ed. USA: Wiley-Blackwell; 2013. 105–111 p.
29. Railliet A, Henry A. Observations sur les strongylides du genre *Nematodirus*. Bull Soc Path Exot. 1912;5(1):35–39. DOI: 10.5962/bhl.part.25474
30. May HG. Observations on the nematode genus *Nematodirus*, with descriptions of new species. Proc U S Natl Mus. 1920;58(2350):577–588. DOI: 10.5479/si.00963801.58-2350.577
31. Shore DA. Differentiation of eggs of nematodes parasitic in domestic ruminants in the United States. Tech Bull. 1939;694:1–11. [available at]
32. Tetley JH. Ecological studies on Nematodirus species in sheep in Manawatu district, New Zealand. J Helminthol. 1935;13(1):41–58. DOI: 10.1017/S0022149X0000331X
33. Tetley JH. Differentiation of eggs of trichostrongylid species *Nematodirus filicollis* and *Nematodirus spathiger*. J Parasitol. 1941;27(6):473. DOI: 10.2307/3272522
34. Kates KC, Turner JH. Life cycle of *Nematodirus spathiger* in sheep and other ruminants. Am J Vet Res. 1955;16(58):105–115. [available at]
35. Thomas RJ. Comparative study of infective larvae of *Nematodirus* species parasitic in sheep. Parasitology. 1957;47(1-2):60–65. DOI: 10.1017/S0031182000021752
36. Soulsby EJL. Helminths, arthropods and protozoa of domesticated animals. 6th ed. UK: Baillière Tindall; 1968:233–234 p. [available at]
37. Lichtenfels JR, Pilitt PA. Cuticular ridge patterns of *Nematodirus* parasitic in domestic ruminants of North America, with a key to species. Proc Helminthol Soc Wash. 1983;50(2):261–274. [available at]
38. Mönnig HO. Diagnosis of nematode infestation in sheep. 17th Rep Dir Vet Serv South Africa. 1931;1:255–264. [available at]
39. Dikmans G, Andrews JS. Comparative morphology of infective larvae of nematodes parasitic in sheep. Trans Am Microsc Soc. 1933;52(1):1. DOI: 10.2307/3222221
40. Van Wyk JA, Cabaret J, Michael LM. Morphological identification of nematode larvae of small ruminants and cattle. Vet Parasitol. 2004;119(4):277–306. DOI: 10.1016/j.vetpar.2003.11.012
41. Van Wyk JA, Mayhew E. Morphological identification of parasitic nematode infective larvae: practical laboratory guide. Onderstepoort J Vet Res. 2013;80(1):539. DOI: 10.4102/ojvr.v80i1.539
42. Melnychuk V, Yevstafieva V, Bakhur T, Antipov A, Feshchenko D. Prevalence of gastrointestinal nematodes in sheep (*Ovis aries*) in Ukraine. Turk J Vet Anim Sci. 2020;44(5):985–993. DOI: 10.3906/vet-2004-54
43. Herlich H. The Life History of *Nematodirus helvetianus* May, 1920, a Nematode Parasitic in Cattle. J Parasitol. 1954;40(1):60–70. DOI: 10.2307/3274257

الجوانب الخاصة للتطور الخارجي لـ Nematodirus spathiger (Nematoda, Molineidae) في درجات الحرارة اليومية

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الخلاصة

تشمل الخصائص البيئية في تطور المراحل الخارجية للديدان الخيطية بعض الجوانب المهمة لبقائها وتوزيعها في عوامل غير حيوية مختلفة. كان الهدف من هذا العمل هو الدراسة المختبرية لبعض الخصائص البيولوجية للمراحل الخارجية لـ *Nematodirus spathiger* المأخوذة من الماشية ونمت في درجات حرارة مختلفة (+٢٠، ٢٣، ٢٦، ٢٩ و ٣٢ درجة مئوية). تم العثور على درجة الحرارة يؤثر بشكل كبير على فترة التطور الخارجي للبيض *N. spathiger*، سواء حتى L3 وعلى المراحل الفردية. تؤثر درجة الحرارة أيضا على بقاء الديدان الخيطية على حد سواء بيض و فقس L3. عند ٣٢ درجة مئوية، كانت معدلات بقاء البيض والفقس L3 هي الأعلى، مع أقصر فترة تطور. عند درجة الحرارة هذه، اكتمل تفقيس L3 بحلول اليوم ١٠، وكان بقائهم على قيد الحياة ٩٤,٣٪. مع انخفاض درجة الحرارة، طال وقت التطور الخارجي ، وكانت معدلات البقاء على قيد الحياة أقل: عند ٢٩ درجة مئوية – ١٢ يوما و ٩١,٩ ٪، عند ٢٦ درجة مئوية – ١٨ يوما و ٨٦,٧ ٪، عند ٢٣ درج مئوية – ٢٢ يوما و ٨٠,٠ ٪، عند ٢٠ درجو مئوية – ٣٠ يوما و ٥٦,٧ ٪ على التوالي. خلال تطوير بيض *N. spathiger* انخفض طول وسمك قشرته بنسبة ١٠,٦ و ٢٢,٦ ٪ على التوالي، وزاد عرضها بنسبة ١٠,٥٪. بالإضافة إلى ذلك، لوحظت تغييرات في المعلمات المورفومترية أثناء النضج L3، وزاد طول ونسبة الفتيل إلى STE بنسبة ٧,١ و ٧,٨ ٪ ، وانخفض عرض الجسم مع الغمد وبدونه ، وانخفض طول STE بنسبة ٤٠,٤ ٪ و ٣٧,٩ ٪ و ٨,٨ ٪ على التوالي.