

Comparative Analysis of the Parasitocenoses of Amphibians from the Family Ranidae (Anura) in the Urbanization Gradient

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Abstract—The parasitofauna of frogs from the fam. Ranidae (*R. arvalis*, *R. temporaria*, and *P. ridibundus*) has been studied in the urbanization gradient. A total of 22 parasitic species belonging to the following four taxonomic groups have been found: Protozoa, Trematoda, Monogenoidea, and Nematoda. The transformation of parasitocenoses with the growth of urbanization along with the preservation of specifics typical for the adaptive subzone in each amphibian species is accompanied with their depletion as well as with an increase in the proportion of trematodes, which results in a decrease in the system stability and increases the probability of parasitic pollution of the environment.

Keywords: parasitofauna, tailless amphibians, urbanization gradient, invasive species, structure of dominance, Ranidae

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INTRODUCTION

Transformation of such coevolutionary systems as parasite–host and changes in their coadaptive relations under the conditions of urbanization and environmental fragmentation are important and demand thorough investigation [1]. It is known that the synergistic effect of pollutants and other anthropogenic factors leads to a functional imbalance of the immune system in amphibians, thereby making them more sensitive to parasitic invasions [2]. If invasive species appear, it increases the potential risk associated with the invasion of new parasitic systems in autochthonous communities. Such changes may lead to outbreaks of zoonotic invasions [3], which considerably affect environmental health. Thus, monitoring of the dynamics of parasite–host relations and parasitic pollution of the environment are very important.

The parasitocenoses of tailless amphibians in urban territories have been studied insufficiently and, as a rule, are based on a single host species [4–9]. Anthropogenic landscapes on the eastern slope of the Middle Urals, including urban territories, are inhabited by three representatives of tailless amphibians from the family Ranidae, including two native species of brown frogs (moor frog—*Rana arvalis* Nilsson, 1842; common frog—*Rana temporaria* Linnaeus, 1758) and one invasive species (march frog—*Pelophylax ridibundus* Pallas, 1771) from the group of green frogs. This study aims at a comparative analysis of the parasitofauna of

the above species in the urbanization gradient, which is clearly of high interest.

MATERIAL AND METHODS

The material for this study was sampled during the period of 2010–2014 from the territory of the urban agglomeration and suburb forest area of Yekaterinburg. The parasitofauna of amphibians was investigated by the method of complete helminthological dissection [10]. A total of 1736 frog specimens were examined: *R. arvalis*—1106; *R. temporaria*—196 (data from the multistory area are not available due to the death of the population); *P. ridibundus*—434.

Parasite identification [11, 12] and infestation assessment were performed based on the following indices: invasion extensity (IE, %)—the proportion of infested host specimens in the studied sample; abundance index of parasites (AI, spec/host)—the average abundance of parasites belonging to a particular species in all host specimens [13]. To assess the significance of differences in IE and AI, the Yates corrected χ^2 test and the analysis of variance were used, respectively. Due to the high abundance of Protozoa and the higher pathogenic importance of macroparasites for amphibians [14], the structure of dominance was analyzed without taking into account protozoans. The dominance was assessed according to A.A. Kirillov [15]: dominants—species comprising 30% and more of the total

number of animals; subdominants—10–30%; common—1–10%; rare—0.1–1%; solitary—0.01–0.1%.

For comparison in the urbanization gradient, urban landscape typification was performed [16]. Four zones inhabited by amphibians were singled out in the large industrial city: multistory area (II), low-rise area (III), forest-park area (IV), and suburb area (C—control). Each habitat was assigned to a particular zone based on the degree of its development by humans (number of stories in the residential area, residential density, and level of household and industrial pollution). As the control group, a suburb population at a distance of 23 km from Yekaterinburg for *R. arvalis* and the populations in the floodplains of the Sysert' and Serga Rivers for *R. temporaria* and a population from Kozhakul' Lake for *P. ridibundus* were used. Based on the mineral composition of surface waters and the temperature regime, a gradient differentiation, which is confirmed annually with the hydrochemical analysis of spawning ponds, is found. Urban water bodies are characterized by a significantly higher ($F = 2.99$, $p = 0.036$) amount of rapidly oxidized organic matter (BOD_5) [16], which is the evidence of considerable eutrophication. The hydrochemical analyzes were carried out in the Laboratory of Physical and Chemical Analysis, Ural State Mining University.

Overlapping of the species spectra for parasites was calculated using the modified Morisita's overlap index [17]:

$$C_M = \frac{2 \sum P_{ix} P_{iy}}{\sum (P_{ix}^2 + P_{iy}^2)},$$

where P_{ix} is the proportion of species i in the spectrum of sample x , P_{iy} is the proportion of species i in the spectrum of sample y . Statistical processing of the results was carried out with the help of the Statistica program package for Windows 6.0.

RESULTS AND DISCUSSION

Species Composition of Parasites in R. arvalis, R. temporaria, and P. ridibundus

A total of 22 parasite species belonging to the following four taxonomic groups were found in the studied representatives of the family Ranidae: two Protozoa, 13 Trematoda, one Monogenea, six Nematoda (geohelminthes). The species composition of parasites and their infestation indices are given in Table 1. Three representatives of the fam. Ranidae have four common parasite species: protozoans—*O. ranarum*, trematodes—*H. cylindracea* and *H. volgensis*, mtc., nematode—*O. filiformis*.

Structure of Parasitic Communities in Three Amphibian Species in the Urbanization Gradient

The composition of a parasitic community depends directly on the ecological niche and life cycle

of its host species. Transformation of biocenotic relations between amphibians under the conditions of urban ecosystems reduces the range of their intermediate and definitive hosts and causes changes in the species spectra of parasites and their proportion and abundance. It is known that the feeding spectra of *R. arvalis* and *R. temporaria* are depleted and specific under the conditions of urbanization [18]. It was shown in [19] that the diet of adult specimens of *R. arvalis* from zone II has no orthopterans and lepidopterans. The analysis of the integration of such an invasive species as *P. ridibundus* in the biocenoses revealed an insignificant overlapping of its feeding spectrum with that of brown frogs [20] along with the high proportion of surface-dwelling food objects (60–75%), where large-sized organisms (Odonata, Carabidae, Hymenoptera), which are the intermediate hosts of some trematodes and not consumed by the aboriginal amphibians, are present [21]. *R. arvalis* and *R. temporaria* live on the ground surface outside the reproduction period; *R. arvalis* overwinters on the ground as well, while *R. temporaria* spends this time at the bottom of water bodies. *P. ridibundus* spends the greater part of its active period of the life cycle in water bodies and is, similarly to *R. temporaria*, a water-overwintering species. Along with the biotic factors, the reduction of the surface part of the habitats of urban amphibian populations and the growth of their local density, temperature increase, and moisture content decrease in soil, and eutrophication of water bodies in the urbanization gradient [22] also influence the transformation of parasitocenoses.

The structure of amphibian parasitocenoses is given in Fig. 1. The proportion of parasitic protozoans in *R. arvalis* varies from 19.38 to 59.78% for the studied territories (see Fig. 1a). Among the two registered opaline species, *O. ranarum* dominates everywhere (from 92.84 to 100%) over *C. dimidiata* (from 1.96 to 7.16%), which is completely absent in zone III. The proportion of trematodes in *R. arvalis* is relatively high in the urban territory.

R. temporaria, as compared to *R. arvalis*, in all the studied territories is characterized by a higher proportion of protozoans represented only by *O. ranarum* (see Figs. 1a, 1b). The high occurrence of *O. ranarum* in brown frogs is determined by the synchronization of mass invasion of this opaline with spawning in *R. arvalis* and *R. temporaria* [23]. The proportion of nematodes in *R. temporaria* with the growth of urbanization decreases from 9.72 and 9.47% in the forest parks and control (respectively) to 2.3% in the residential zone, being always significantly higher than the proportion of trematodes. It is known that the parasitofauna of *R. temporaria* is prevailed by geohelminthes due to the high moisture content in the biotope [24]. It was shown that *R. temporaria* is confined to cold subsurface water outlets [25], which are not preferred by trematodes and inhibit the development of their eggs [26]. Solitary specimens of the monogenean *P. integerrimum*

Table 1. Species composition of parasites and the infestation rates of the studied amphibian species

Parasite species	Amphibian species in zones											
	<i>R. arvalis</i>				<i>R. temporaria</i>				<i>P. ridibundus</i>			
	II	III	IV	C	II	III	IV	C	II	III	IV	C
Protozoa												
<i>Opalina ranarum</i> Dujardin, 1841	$\frac{45.6}{6.08}$	$\frac{47.3}{7.03}$	$\frac{50.1}{5.67}$	$\frac{45.2}{17.13}$	–	$\frac{48.1}{12.2}$	$\frac{46.8}{17.1}$	$\frac{50.0}{38.5}$	$\frac{6.1}{1.8}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{1.3}{1.7}$
<i>Cepedea dimidiata</i> (Metcalf, 1923)	$\frac{0.9}{0.47}$	$\frac{0}{0}$	$\frac{1.0}{0.20}$	$\frac{0.5}{0.34}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{68.7}{46.6}$	$\frac{69.8}{53.4}$	$\frac{60.2}{35.4}$	$\frac{70.0}{70.7}$
Trematoda												
<i>Dolichosaccus rastellus</i> (Olsson, 1876)	$\frac{33.8}{1.49}$	$\frac{1.2}{0.02}$	$\frac{2.0}{0.05}$	$\frac{1.4}{0.03}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0.9}{0.009}$	$\frac{0}{0}$	$\frac{7.5}{2.2}$
<i>Opisthioglyphe ranae</i> (Frölich, 1791)	$\frac{5.3}{0.18}$	$\frac{6.0}{0.11}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{6.1}{10.3}$	$\frac{14.7}{12.4}$	$\frac{0}{0}$	$\frac{46.3}{8.9}$
<i>Pleurogenes claviger</i> (Rudolphi, 1819)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{1.6}{0.11}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{2.5}{0.1}$
<i>Pleurogenes intermedius</i> Issaitschikov, 1926	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{1.3}{0.13}$
<i>Prosotocus confusus</i> (Looss, 1894)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{4.3}{19.0}$	$\frac{13.8}{17.4}$	$\frac{4.1}{22.3}$	$\frac{3.8}{1.2}$
<i>Pleurogenoides medians</i> (Olsson, 1876)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{1.7}{0.09}$	$\frac{0}{0}$	$\frac{0}{0}$
<i>Pleurogenoides stromi</i> (Travassos, 1930)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{1.3}{0.01}$
<i>Gorgoderina skrjabini</i> Pigulewsky, 1953	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{1.3}{0.01}$
<i>Gorgoderia pawlowskyi</i> (Pigulewsky, 1952)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{2.4}{0.2}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$
<i>Haplometra cylindracea</i> (Zeder, 1800)	$\frac{0.4}{0.04}$	$\frac{0}{0}$	$\frac{4.4}{0.14}$	$\frac{0.5}{0.005}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{18.3}{0.6}$	$\frac{0}{0}$	$\frac{3.4}{0.04}$	$\frac{0}{0}$	$\frac{3.8}{0.09}$
<i>Pneumonoeces variegatus</i> (Rudolphi, 1819)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{5.0}{0.2}$
<i>Echinoparyphium recurvatum</i> (Linstow, 1873) mtc	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{1.2}{0.09}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$
<i>Holostephanus volgensis</i> (Sudarikov, 1962) mtc	$\frac{25.0}{6.26}$	$\frac{42.5}{29.0}$	$\frac{26.9}{0.4}$	$\frac{36.7}{11.5}$	–	$\frac{3.8}{0.08}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0.9}{0.02}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$
Monogenoidea												
<i>Polystoma integerrimum</i> (Frölich, 1791)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	–	$\frac{0}{0}$	$\frac{3.2}{0.03}$	$\frac{1.2}{0.01}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$
Nematoda												
<i>Oswaldocruzia filiformis</i> (Goeze, 1782)	$\frac{8.3}{0.18}$	$\frac{6}{0.16}$	$\frac{13.6}{0.58}$	$\frac{4.3}{0.08}$	–	$\frac{3.8}{0.25}$	$\frac{41.9}{1.26}$	$\frac{37.8}{1.89}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0.8}{0.008}$	$\frac{5.0}{0.006}$
<i>Cosmocerca ornata</i> (Dujardin, 1845)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{4.2}{0.07}$	$\frac{4.3}{0.08}$	–	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{1.2}{0.07}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$

Table 1. (Contd.)

Parasite species	Amphibian species in zones											
	<i>R. arvalis</i>				<i>R. temporaria</i>				<i>P. ridibundus</i>			
	II	III	IV	C	II	III	IV	C	II	III	IV	C
<i>Neoraillietnema praeputiale</i> (Skrjabin, 1916)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0.6}{0.006}$	$\frac{0.5}{0.01}$	—	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{6.1}{0.11}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$
<i>Aplectana acuminata</i> (Schränk, 1788)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	—	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{2.4}{0.22}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$
<i>Neoxysomatium brevicaudatum</i> (Zeder, 1800)	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	—	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{2.4}{0.22}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$
<i>Rhabdias bufonis</i> (Schränk, 1788)	$\frac{1.3}{0.02}$	$\frac{0}{0}$	$\frac{2.6}{0.08}$	$\frac{0.5}{0.06}$	—	$\frac{1.9}{0.04}$	$\frac{11.3}{0.6}$	$\frac{22.0}{1.6}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$	$\frac{0}{0}$
Number of parasite species	8	5	10	9	—	4	5	10	5	6	3	12

Above line—invasion extensity (%); under line—abundance index (spec/host); mtc—metacercariae.

were found in the populations of *R. temporaria* from zone IV and control.

The suburb population of *P. ridibundus* is characterized by a higher, if compared to the urban territories, proportion of protozoans (see Fig. 1c), which are represented by two species. However, in contrast to brown frogs, *P. ridibundus* spawning at a later time is dominated by *C. dimidiata* (from 96.21 to 100%). For the urban populations of *P. ridibundus*, a higher proportion of trematodes in the urban lands was registered, while nematodes were represented by solitary specimens of *O. filiformis* in the populations of zone IV and control.

Due to the complex life cycles and specific hosts, amphibian macroparasites are of potential interest as the indicators of transformations in ecosystems. In the complex balanced community with high species diversity, the pathogenicity of parasites is low [27, 28]. The synergic effect of pollutants leads to the functional disbalance in the immune system of amphibians, thereby making them more prone to parasitic invasions [2]. Under these conditions, amphibian helminths can have a direct negative influence on their hosts [14]. Therefore, we analyzed the structure of macroparasitic complexes in more detail. In Table 2, data are provided on the ratio between the numbers of macroparasitic species in the studied amphibians in the urbanization gradient.

The dominant parasitic species in *R. arvalis* (without regarding protozoans) for all territories is the trematode *H. volgensis*, mtc., which infests the host as a result of active percutaneous penetration of cercariae in the host body. The superdominance of *H. volgensis* larvae was registered in zone III and control (see Tables 1, 2). The death of *R. arvalis* yearlings full with cysts of this parasite was repeatedly registered (Fig. 2). The highest diversity of nematodes was observed in the suburb populations of *R. arvalis* and zone IV: *O. fili-*

formis, *C. ornata*, *R. bufonis*, *N. praeputiale* (see Table 2). In zone IV, *O. filiformis* becomes a common species at the highest infestation parameters in the urbanization gradient (see Table 1). In the residential zones, the geohelminths *C. ornata* and *N. praeputiale* disappear. At the same time, the trematode *O. ranae* was registered only for these territories. The proportion of and infestation with *D. rastellus*, whose intermediate hosts are gastropods of the genus *Lymnaea*, increase significantly in zone II (see Tables 1, 2). *R. arvalis* acts as the definitive and additional, i.e., amphixenic, host for *D. rastellus* [21]. The accumulative character of urban lands favors an increase in eutrophication (BOD₅ and mineralization) of the spawning pods of amphibians [29], algal biomass gain, increase in the abundance of intermediate hosts (gastropods, insects) and the abundance of cercariae produced by them. This increases the chances of trematode invasion [30], which enhances in *R. arvalis* from the urban territories.

The helminthofauna of the suburb populations of *R. temporaria* and the populations of zone IV is dominated by nematodes (see Table 1, 2). The core is made by the geohelminths *O. filiformis* and *R. bufonis*. The following species are commonly found: *C. ornata*, *N. praeputiale*, *A. acuminata*, and *N. brevicaudatum*. In the forest-park and suburb zones, *P. integerrimum*, a highly specified monogenean with the restricted range of hosts, was found [14]. It was shown [31] that representatives of the class Monogenoidea are sensitive to increased water mineralization, which may explain their absence in the urban populations of *R. temporaria*.

In the urban territories, the species composition of parasites is depleted, which is accompanied by changes in the degree of dominance among the remaining species: the proportion of *R. bufonis* is reduced, the proportion of *O. filiformis* (with the low infestation rates as compared to other zones) from

fam. Trichostrongyloidea that has a coating resistant to mineral and organic substances [32], which, probably, determines the high proportion of this species in the residential zone (zone III), grows. Along with *R. bufonis*, the trematode *H. volgensis*, mtc. becomes a subdominant. The absence of *C. ornata* in brown frogs from the residential territories must be associated with the sensitivity of larvae of this nematode to the pollution level in the urban environment. Similar trends (reduction in the species and taxonomic diversity, decrease in the invasion extensity and intensity) in the urban parasitocenoses of *R. temporaria* were registered by other researchers [4, 7]. The infestation of amphibians with *R. bufonis* nematodes occurs through their percutaneous penetration (at the stage of invasive larvae) from the humid soil and intensifies in the rain season [33]. Thus, the stenotopic and hydrophilous *R. temporaria* is more invaded with nematodes as compared to the eurytopic *R. arvalis*, which uses eutrophic temporary water bodies for its reproduction, where the chances of infestation with trematodes increase [34].

The parasitic complexes of *P. ridibundus*, which has been an invader on the eastern slope of the Middle Urals since the second half of the 20th century, are of particular interest. The parasitocenoses of the native populations of *P. ridibundus* in the Central Black Earth region include 21 trematode species and five nematode species [35]. On the western slope of the Southern Urals, they comprise up to 16 trematode species and three nematode species [8]. The parasitofauna of the studied territories is characterized by the dominance of trematodes with various localization, while nematodes are found as solitary specimens (see Tables 1, 2).

The helminthocenosis of the suburb population of *P. ridibundus* includes ten species, six of which (*P. claviger*, *P. stromi*, *P. variegatus*, *P. intermedius*, *D. rastellus*, *G. skrjabini*) were not registered in the urban territories. Among helminths, *O. ranae* is the dominant and *D. rastellus* is the subdominant. In the youngest population (formed in 1998) of *P. ridibundus* in zone IV, one mass species, *P. confusus*, was revealed and the solitary finding of *O. filiformis*, the infestation by which must be accidental, was made.

In the populations of the residential territories, the parasitofauna of *P. ridibundus* is considerably depleted as compared to the control and is represented mainly by *P. confusus* and *O. ranae*, i.e., these are adult forms of trematodes, for which amphibians are the definitive host (autogenous biohelminths): *O. ranae* is transmitted with food objects (gastropods, tadpoles), while *P. confusus* has a flexible life cycle [36] and comprises a higher proportion in the helminthocenoses of the urban populations of *P. ridibundus* due to the wide range of intermediate and definitive hosts. Furthermore, *P. ridibundus* in zone II had solitary *H. volgensis* metacercariae. The high proportion of trematodes in the parasitocenosis of *P. ridibundus* is explained by the specifics of ecophysiology: its hemoglobin is charac-

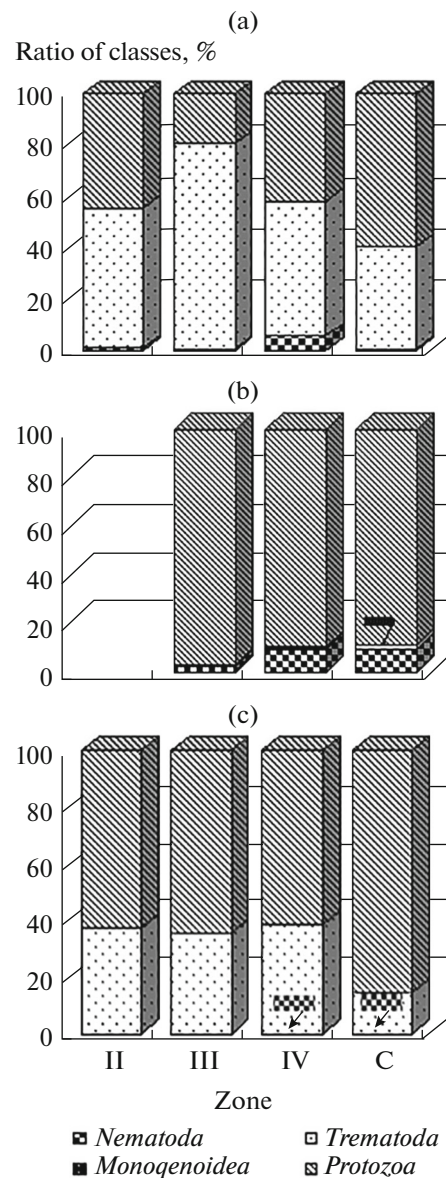


Fig. 1. Ratio of parasite classes in the studied specimens in the urbanization gradient: (a) *R. arvalis*, (b) *R. temporaria*, (c) *P. ridibundus*.

terized by the high affinity for oxygen [37], which allows this species to live and overwinter in eutrophic water bodies of the urban territories (in contrast to *R. temporaria*), where the abundance of intermediate hosts of trematodes (limneids) and temperatures (by 3–5°C) higher than in the control [29] ensure favorable conditions for the mass emergence of cercariae. The total absence of *C. ornata* in *P. ridibundus* is explained by the fact that the invasion of amphibians by this nematode is not registered in the case of low infestation of natural populations, which was demonstrated by the previous research performed on the populations of *P. ridibundus* within the native habitat [8]. We associate the relatively poor composition of

Table 2. Ratio of parasite species (%) in the studied amphibian species in the urbanization gradient

Parasite species	Amphibian species in zones											
	<i>R. arvalis</i>				<i>R. temporaria</i>				<i>P. ridibundus</i>			
	II	III	IV	C	II	III	IV	C	II	III	IV	C
	Trematoda											
<i>Dolichosaccus rastellus</i> (Olsson, 1876)	18.33	0.06	0.59	0.28	—	0	0	0	0	0.03	0	16.88
<i>Opisthioglyphe ranae</i> (Frölich, 1791)	2.21	0.37	0	0	—	0	0	0	35.24	41.39	0	69.66
<i>Pleurogenes claviger</i> (Rudolphi, 1819)	0	0	0	0	—	0	5.65	0	0	0	0	0.78
<i>Pleurogenes intermedius</i> Issaitschikov, 1926	0	0	0	0	—	0	0	0	0	0	0	0.97
<i>Prototocus confusus</i> (Looss, 1894)	0	0	0	0	—	0	0	0	64.7	58.15	99.96	8.97
<i>Pleurogenoides medians</i> (Olsson, 1876)	0	0	0	0	—	0	0	0	0	0.29	0	0
<i>Pleurogenoides stromi</i> (Travassos, 1930)	0	0	0	0	—	0	0	0	0	0	0	0.1
<i>Gorgoderina skrjabini</i> Pigulewsky, 1953	0	0	0	0	—	0	0	0	0	0	0	0.1
<i>Gorgodera pawlowskyi</i> (Pigulewsky, 1952)	0	0	0	0	—	0	0	0	0	0	0	0
<i>Haplometra cylindracea</i> (Zeder, 1800)	0.05	0	1.78	0.04	—	0	0	0	0	0.14	0	0.68
<i>Pneumonoeces variegatus</i> (Rudolphi, 1819)	0	0	0	0	—	0	0	0	0	0	0	1.37
<i>Echinoparyphium recurvatum</i> (Linstow, 1873) mtc	0	0	1.14	0	—	0	0	0	0	0	0	0
<i>Holostephanus volgensis</i> (Sudarikov, 1962) mtc	76.98	99.04	87.35	97.73	—	21.05	0	0	0.06	0	0	0
	Monogenoidea											
<i>Polystoma integerrimum</i> (Frölich, 1791) <i>integerrimum</i>	0	0	0	0	—	0	1.61	0.25	0	0	0	0
	Nematoda											
<i>Oswaldocruzia filiformis</i> (Goeze, 1782)	2.16	0.53	7.23	0.69	—	68.42	62.9	38.65	0	0	0.04	0.49
<i>Cosmocerca ornata</i> (Dujardin, 1845)	0	0	0.89	0.69	—	0	0	1.5	0	0	0	0
<i>Neoraillietema praeputiale</i> (Skrjabin, 1916)	0	0	0.08	0.08	—	0	0	2.24	0	0	0	0
<i>Aplectana acuminata</i> (Schrank, 1788)	0	0	0	0	—	0	0	4.49	0	0	0	0
<i>Neoxysomatium brevicaudatum</i> (Zeder, 1800)	0	0	0	0	—	0	0	4.49	0	0	0	0
<i>Rhabdias bufonis</i> (Schrank, 1788)	0.27	0	0.94	0.49	—	10.53	29.84	32.67	0	0	0	0
Number of parasites	1855	4885	4038	2469	—	19	124	401	3371	3465	2749	1025

mtc—metacercariae.

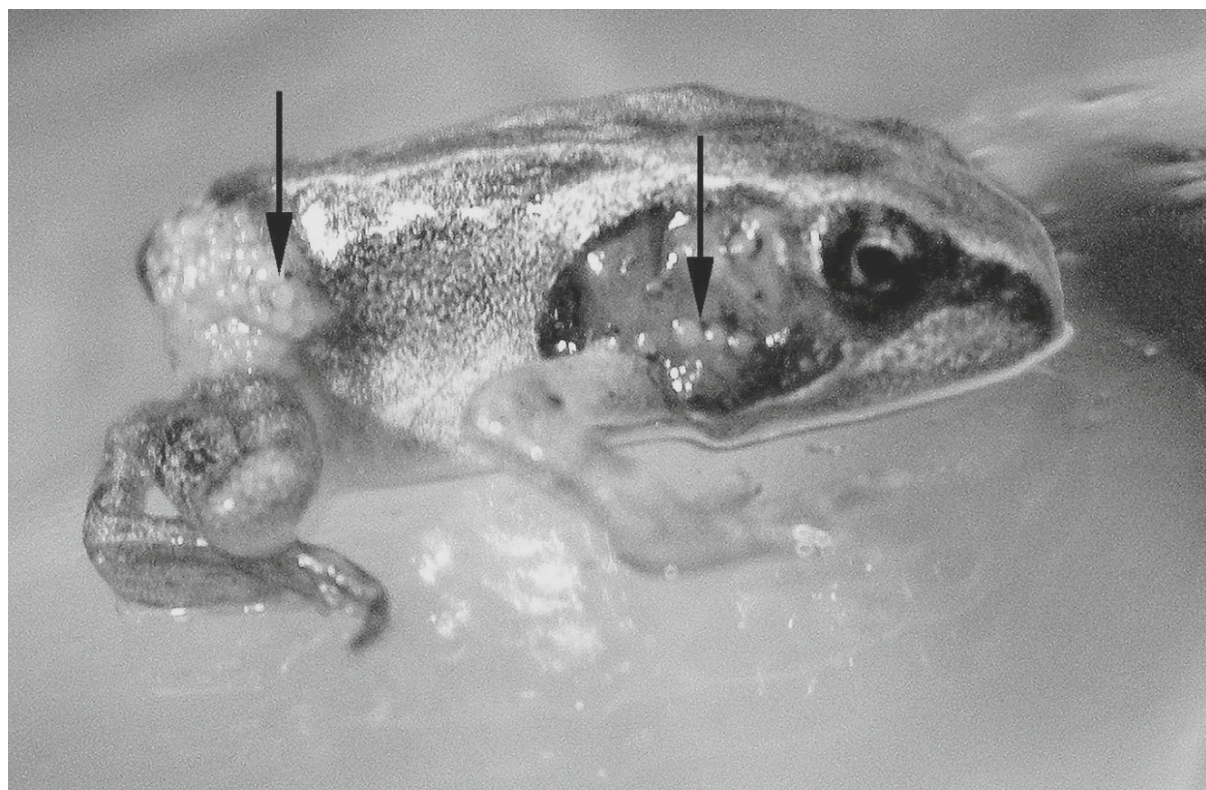


Fig. 2. Dead yearling of *R. arvalis* infested by *Holostephanus volgensis* metacercariae (the forest-park zone of Yekaterinburg; photo by V.L. Vershinin).

parasitic communities in *P. ridibundus* from the studied populations with the fact that this frog is an invasive species in the territory under consideration.

Despite the anthropogenic transformation of habitats of amphibians, the spectra of parasitic communities are considerably similar to their initial state. There was a significant overlapping of the spectra based on the Morisita's index for all the investigated Ranidae species in the urbanization gradient between the zones: 72.9–97.8% for *R. arvalis*, 99.5–99.9% for *R. temporaria*, 83.8–99.6% for *P. ridibundus*. At the same time, the comparison of the summarized spectra of parasitofauna (Table 2) showed their similarity in brown frogs (57%) along with the significant difference with *P. ridibundus*: 3% similarity with *R. arvalis* and 1% with *R. temporaria*.

Changes in Infestation Rates in the Urbanization Gradient

Data on the infestation of amphibians with parasites of various taxonomic groups are given in Table 3. There were no intraspecific differences in IE of Protozoa in all studied Ranidae in the urbanization gradient. This parameter is significantly higher in *P. ridibundus* as compared to *R. arvalis* ($\chi^2 = 14.15$, $p = 0.0002$) and *R. temporaria* ($\chi^2 = 6.79$, $p = 0.009$) in the control and

residential territories. The abundance index of protozoans in the studied amphibian species is significantly higher than in the suburb territory: ($F(3, 1102) = 21.895$, $p \leq 0.0001$ for *R. arvalis*, $F(2, 193) = 7.0970$, $p = 0.001$ for *Rana temporaria*, and $F(3, 430) = 3.0578$, $p = 0.028$ for *P. ridibundus*). The lowest AI of Protozoa was registered in *R. arvalis*, the most terrestrial species, while the highest in *P. ridibundus*. Based on the infestation of *R. arvalis* with trematodes, there was registered a significant increase in IE in the residential territories ($\chi^2 = 6.77$, $p = 0.009$ and $\chi^2 = 4.08$, $p = 0.043$ for zones II and III as compared to the control, respectively; for the same zones as compared to zone IV $\chi^2 = 18.15$, $p = 0.0001$ and $\chi^2 = 11.37$, $p = 0.007$, respectively). The AI is significantly higher ($F(3, 1102) = 13.655$, $p \leq 0.00001$) in zone III, where the BOD₅ is maximum [29].

The extensity of invasion of *R. arvalis* by nematodes is higher in zone IV as compared to other territories ($\chi^2 = 4.32$ – 8.61 , $p = 0.003$ – 0.039); the AI for this zone is also reliably higher ($F(3, 1102) = 7.7838$, $p = 0.00004$). The infestation of *R. temporaria* by multicellular parasites decreases in the urbanized territories. The IE by trematodes is significantly lower in all urban territories as compared to the control ($\chi^2 = 4.76$, $p = 0.029$ and $\chi^2 = 8.33$, $p = 0.004$ for zones III and IV, respectively). The AI decreases reliably as compared to the suburb

Table 3. Infestation of the studied amphibian species by various parasite groups in the urbanization gradient

Taxon	Infestation indicator	Zone											
		II			III			IV			C		
		R. a.	R. t.	P. r.	R. a.	R. t.	P. r.	R. a.	R. t.	P. r.	R. a.	R. t.	P. r.
Protozoa	IE	$\frac{46.49}{r}$	—	$\frac{72.17}{a}$	$\frac{47.31}{r}$	$\frac{48.07}{r}$	$\frac{69.82}{a,t}$	$\frac{51.09}{r}$	$\frac{46.77}{r}$	$\frac{60.16}{a,t}$	$\frac{45.71}{r}$	$\frac{50.0}{r}$	$\frac{71.25}{a,t}$
	AI	$\frac{6.55}{4,r}$	—	$\frac{48.41}{4,a}$	$\frac{7.03}{4,t,r}$	$\frac{12.15}{4,a,r}$	$\frac{53.37}{3,a,t}$	$\frac{5.87}{4,r}$	$\frac{17.08}{4,a,r}$	$\frac{35.36}{2,4,a,t}$	$\frac{17.47}{1,2,3,t,r}$	$\frac{38.52}{2,3,a}$	$\frac{72.3}{1,3,a}$
	IE	$\frac{50.44}{3,4,r}$	—	$\frac{11.30}{2,3,4,a}$	$\frac{48.50}{3,4,t,r}$	$\frac{3.84}{4,a,r}$	$\frac{30.17}{1,3,4,t,a}$	$\frac{33.33}{1,2,t,r}$	$\frac{1.61}{4,a}$	$\frac{4.06}{1,2,4,a}$	$\frac{37.61}{1,2,t,r}$	$\frac{18.29}{2,3,a,r}$	$\frac{58.75}{1,2,3,a,t}$
Trematoda	AI	$\frac{7.94}{2,r}$	—	$\frac{29.31}{a}$	$\frac{29.09}{1,3,4,t}$	$\frac{0.07}{4,a,r}$	$\frac{29.87}{t}$	$\frac{7.32}{2,t}$	$\frac{0.11}{4,a,r}$	$\frac{22.34}{t}$	$\frac{11.52}{2,t}$	$\frac{0.76}{2,3,a,r}$	$\frac{12.75}{t}$
	IE	—	—	—	—	—	—	—	3.22	—	—	1.21	—
Monogeneoidea	AI	—	—	—	—	—	—	—	0.03	—	—	0.01	—
	IE	$\frac{9.21}{3}$	—	—	$\frac{5.99}{3}$	$\frac{5.76}{3,4}$	—	$\frac{15.17}{1,2,4,t}$	$\frac{45.16}{2,a,r}$	$\frac{0.81}{t}$	$\frac{9.05}{3,t}$	$\frac{48.78}{2,a,r}$	$\frac{5.0}{t}$
Nematoda	AI	$\frac{0.19}{3}$	—	—	$\frac{0.16}{3}$	$\frac{0.28}{3,4}$	—	$\frac{0.74}{1,2,4,r}$	$\frac{1.85}{2,4,a,r}$	$\frac{0.008}{a,t}$	$\frac{0.23}{3,r,t}$	$\frac{4.10}{2,3,a,r}$	$\frac{0.063}{a,t}$
	N	228	—	115	167	52	116	501	62	123	210	82	80

R. a.—*R. arvalis*; R. t.—*R. temporaria*; P. r.—*P. ridibundus*; IE—invasion extensity (%); AI—abundance index (spec/host); significant differences: above line—value; under line—significance (inside the amphibian species between zones: 1—with zone I; 2—with zone II; 3—with zone III; 4—with zone IV; 4—with zone C; between the amphibian species inside the zone: a—with *R. arvalis*; t—with *R. temporaria*; r—with *P. ridibundus*); N—number of amphibian specimens.

territory ($F(2, 193) = 3.6859, p = 0.027$). There are no significant differences between frogs in zone IV and control in the infestation by nematodes, while amphibians in the residential territory are less infested with geohelminths. In zone III, the IE is significantly lower ($\chi^2 = 20.22, p < 0.0001$ as compared to zone IV and $\chi^2 = 25.08, p < 0.001$ as compared to the control); the AI is significantly lower for the residential territory ($F(2, 193) = 9.4209, p = 0.0001$). The analysis of infestation of *P. ridibundus* showed that the proportion of specimens invaded by trematodes (IE) is significantly higher in the control (as compared to zone IV: $\chi^2 = 73.23, p < 0.0001$; zone III: $\chi^2 = 14.74, p = 0.0001$; zone II: $\chi^2 = 47.66, p < 0.0001$), while the AI is higher in the urban territories. Geohelminths, which are low-abundant in the control and zone IV, are completely absent in *P. ridibundus* from the residential part of the city.

CONCLUSIONS

Therefore, all studied species of the family Ranidae are generally infested by protozoans and trematodes. Nematodes were registered in *R. arvalis* and *R. temporaria* in the populations of all zones. They are not found in *P. ridibundus* from the residential territories. The observed change in the ratio of parasites belonging to various taxa in the populations of *R. arvalis*, *R. temporaria*, and *P. ridibundus* is determined by the specifics of the life cycle of their hosts and the differences in the biology of parasitic groups. Each of the studied host species in the suburb habitats has originally a certain variant of the parasitic community, which corresponds to their ecological niche and the microbiotic features of habitats. The parasitocenoses of *R. arvalis*, an eurytopic and the most terrestrial species, is characterized among all studied amphibians by the lowest infestation by protozoans, low infestation with nematodes along with their high original diversity, and high infestation by trematodes. The specific feature of parasitocenoses in *R. arvalis* within the studied territory is the high, as compared to the other two species, invasion with metacercariae of the trematode *H. volgensis*, which is due, as we assume, to the synchronization of the period of emergence of cercariae and tadpoles of *R. arvalis*. The life cycles of *P. ridibundus* and *R. temporaria* larvae based on the temporal and ecological parameters do not coincide with the cyclicity of *H. volgensis* and, thus, their infestation by this trematode is much lower.

The parasitocenoses of such a stenotopic species as *R. temporaria*, which prefers moist surface biotopes and cold underground water outlets, are characterized, besides the high proportion of Protozoa, by the high diversity of geohelminths and the relatively low proportion of trematodes, as well as the presence of the monogenean registered only in *R. temporaria*. *P. ridibundus*, which is predominantly aquatic, is characterized by the highest infestation by protozoans

along with the high species diversity of trematodes and the occurrence of nematodes as solitary specimens.

The species richness and a number of parameters characterizing the state of parasitic communities for both multicellular parasites and Protozoa in all studied representatives of the family Ranidae change with the growth of urbanization in accordance with transformations in the chorologic structure of amphibian populations and the microbiotopical specifics of urban habitats: the ratio of their aquatic and ground parts, temperature and chemical composition of the environment, host population density, its position in urban cenoses. Along with the general trends expressed in the disappearance of a number of species and the structural simplification of parasitic communities associated with the falling of some intermediate hosts and the depletion of amphibian diet, the process of their transformation occurs within the species specifics. The adaptive subzone of each host canalizes this process in a certain way. The abundance of macroparasites in *R. arvalis* and *P. ridibundus* in the urban territories increases along with the decrease in their diversity. The structural simplification of parasitocenoses in the urbanization gradient and the increase in the proportion of macroparasites at the expense of Trematoda reduces the coadaptive system homeostatis and increases the risks of parasitic pollution of the environment.

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