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SHORT COMMUNICATIONS

Morphological Anomalies in *Rana arvalis* Nilsson, 1842 from Subarctic Urban Areas

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The moor frog *Rana arvalis* Nilsson, 1842 is one of the most ecologically flexible, eurytopic species in Eurasia, with its range extending from the English Channel to Transbaikalia and from Kazakhstan to the Subarctic. In the Urals, this species is almost ubiquitous and occurs in both natural and anthropogenically transformed habitats [1–4]. The adaptive capacity of *R. arvalis* frogs living in the Far North "...reaches amazing perfection: even at about 0°C their digestive enzymes remain highly active, and the amplitude of heart contractions and the rate of ventricular tension development increase" [5]. Such an adaptive potential is largely related to specific features of polymorphism in this species [6–9].

Research on the population features of amphibians inhabiting the Subarctic was initiated in the 20th century [10], but deviant forms of morphogenesis and other qualitative indices characterizing specific features of population genetic structure have not yet been analyzed. The purpose of this study was to perform comparative analysis of the phenological (morphological) pattern of a new *R. arvalis* generation from urban areas of the Subarctic and Middle Urals.

The material was collected in the city of Labytnangi (the Polar Urals) in the zone of multistory buildings on Novaya St. on July 26–27, 2016. The total sample consisted of 69 postmetamorphic (juvenile) *R. arvalis* frogs, which were analyzed for qualitative and quantitative morphological parameters. Body length was measured with a digital caliper (Kraftool, Germany) to an accuracy of 0.01 mm; body weight, with a digital balance (Shimadzu, Japan) to an accuracy of 10 mg.

Two traits were used to analyze specific features of population genetic structure. The first was the frequency of the striata morph [11] phenotypically manifested as the presence of a light dorsomedial stripe extending to the tip of the snout. It is determined by the dominant allele of the diallelic autosomal gene *striata* with complete dominance [12]. Specific features of inheritance make this trait a good marker of changes in the genetic structure of population. The second trait was the frequency of iris depigmentation, which is determined by a recessive mutation [13] whose expressivity and penetrance depend on the temperature during early stages of ontogeny [14].

Identification and analysis of exterior morphological anomalies were performed according to relevant classifications [15, 16] and also using original approaches [17]. The material for comparison was taken from the database for the territory of Yekaterinburg [18] in 1980 to 2013. Within this urban agglomeration, four zones with different habitat conditions for amphibians were distinguished [1]: (I) city center (not inhabited by amphibians) with multistory buildings, asphalt pavement, water bodies highly polluted by industrial emissions, and tubed small rivers and streams; (II) multistory districts with areas under development, wastelands, areas with bare soil, and highly polluted small water bodies; (III) low-rise districts of private houses with orchards and vegetable gardens, parks, and wasteland areas; (IV) park forest zone with multistory districts, areas under development, wastelands, areas with bare soil, and highly polluted small water bodies; habitats in this zone are mainly affected by recreational activities. The control sample (K) was from suburban populations living in a *Sphagnum*–horsetail pine forest 23 km from Yekaterinburg. The habitat in Labytnangi was also classified as a zone of multistory buildings. The acceptability of this typification is confirmed every year by hydrochemical analysis [18].

Statistical data processing was performed with Statistica for Windows 6.0.

Comparative morphological analysis of the sample from Labytnangi revealed no significant differences in body length, weight, and body condition from these parameters in juveniles from Yekaterinburg (Table 1),

Table 1. Morphological parameters of postmetamorphic *R. arvalis* frogs

Habitat (city)	Zone	*L*, mm	*P*, mg	*P*/*L* (body condition)	Number of animals
Labytnangi	II	16.4 ± 0.3	430.0 ± 22.6	25.2 ± 0.85	69
Yekaterinburg	II	16.4 ± 0.07	445.7 ± 4.9	26.7 ± 0.2	1561
Yekaterinburg	III	15.08 ± 0.09	345.1 ± 6.2	22.7 ± 0.3	999
Yekaterinburg	IV	15.02 ± 0.05	385.7 ± 3.7	22.5 ± 0.2	2844
Yekaterinburg	K	14.9 ± 0.07	368.8 ± 4.9	23.7 ± 0.2	1565

while the frequency of striata morph proved to be significantly higher than in juveniles from zone II in Yekaterinburg: 62.3 vs. 42.9% ($\chi^2 = 12.88$; $p = 0.0003$). The total frequency of animals with morphological anomalies was also significantly higher in Labytnangi than in the multistory zone of Yekaterinburg: 21.7 vs. 6.4% ($\chi^2 = 25.65$; $p \ll 0.0001$). The sample from Labytnangi was characterized by a high frequency of morphological anomalies (26.09%). In a decreasing order of occurrence, they include ectrodactyly, iris depigmentation, oligodactyly, abnormal skin pigmentation, and ectromelia (Table 2). For comparison: in the Middle Urals, the proportion of individuals with morphological anomalies among *R. arvalis* juveniles is 1.5–2% in forest populations versus 6–8% in urbanized areas. Iris depigmentation is a recessive mutation occurring with a frequency of about 0.39% (with a maximum of 3%) in forest populations and 1.7% (with a maximum of 7.5%) in urban populations; in the sample from Labytnangi, it was expressed bilaterally in 80% of mutants.

On the whole, a high total frequency of morphological anomalies was revealed among juveniles of new generation from *R. arvalis* population of the study area. Iris depigmentation accounts for a major proportion of all anomalies (33.3%), which is evidence for high homozygosity of this population. As shown for urban populations of amphibians [19–21], fragmentation of habitats and separation of populations into small isolates under the impact of urbanization lead to increasing homozygosity and higher rate of gene pool transformation due to genetic drift accompanied by inbreeding.

Table 2. Frequencies of morphological anomalies (%) in postmetamorphic *R. arvalis* from zones of multistory buildings

Parameter	City	
	Labytnangi	Yekaterinburg
Total number of animals	69	1561
Iris depigmentation	7.24	1.72
Abnormal skin pigmentation	2.9	0.94
Ectromelia	1.45	0.25
Ectrodactyly	11.6	0.25
Oligodactyly	2.9	0.16

The overall frequency of skeletal anomalies, which can be associated with trematode infestation [22], reaches 15.95%. However, only two species of intestinal parasites were found in juveniles of new generation from Labytnangi: nematodes *Oswaldocruzia filiformis* Goeze, 1782 and parasitic protists *Opalina ranarum* Dujardin, 1841. Skeletal anomalies observed in *R. arvalis* juveniles mainly included reduction of distal limb elements in different variants, probably of heterochrony type (Table 2). These results are preliminary and need verification in further studies.

A considerable proportion of striata morph revealed in *R. arvalis* population is evidence for the prevalence of individuals with genetically determined high reactivity of the hematopoietic system, several other physiological processes [6, 7, 9], rapid sexual maturation, and short life span [23]. It appears that the physiological specificity of this morph accounts for an increase in its frequency as an adaptive response of the population to anthropogenic transformation of the environment and to its geochemical background [24, 25] under conditions of strictly limited duration of the active life cycle phase in the Subarctic.

Thus, the results of this study show that *R. arvalis* population from the multistory zone of Labytnangi is characterized by a high level of specificity, which is probably due to insularization of the range and formation of isolates under the impact or urbanization combined with climatic features of the Subarctic.

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