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## *PHENETIC ANALYSIS OF THE ANIMAL POPULATION IN THE URBAN ECOSYSTEM*

*ABSTRACT. In the given work the integrated influence of the factors of urbanization on the phenetic structure of populations of firebugs (Pyrrhocoris apterus L.) and small mammals — red-backed voles (Clethrionomys rutilus P.) and common shrew (Sorex araneus L.) is analyzed. In the populations of firebugs in Ishim the increase in phenetic variability and, accordingly, the frequency of the development of rare phenes of the drawings on their backs according to the increase of anthropogenic load is shown. The population groups of small mammals, living on the territory of a forest park within the city boundaries, differ from the control ones by the frequencies of phenes of non-metrical characters of skulls. The integrated influence of the factors of urbanization leads to strengthening of intraspecific differentiation of populations of small mammals. Phenetic distances (MMD) between the selections of small mammals from the forest park and the relic habitat not far from the city (16 kilometers), calculated according to the complex of 25 characters for the red-backed voles and 9 characters for the common shrew, twice exceed the level defined for rather isolated intrapopulation settlements of the red-backed vole within the continuous area and correspond to the level of the nearest populations isolated by a 30-40-km landscape and geographical barrier.*

*KEY WORDS. Phene, phenetic structure, population, variability.*

A population is always dependent on its environment. In the whole course of life of a population the natural selection tends to preserve those genotypes which lead to formation of the phenotypes which are most suitable for the set of external conditions prevailing in the given time period in the given place. Under the growing anthropogenic burden the role of biological monitoring, characterizing the health condition of the populations and living communities under the cumulative impact of different factors and, through them, the overall condition of the environment, increases. For this purpose attention to the most universal parameters of the functioning of the living systems – variability and development – is paid [7].

One of the indicators of unfavorable ecological condition is increased phenotypic variability. For a number of decades scientists have been studying variability of the wing case pattern of the two-spot ladybird (*Adalia bipunctata*), the nature of industrial melanism of the peppered moth (*Biston betularia*) in England, variability of non-metric skull features of small mammals, etc. The genetic nature of phenotypic variability has been revealed and a hypothesis on the influence of climatic conditions and anthropogenic factors on the phenotypic variability has been made [3]. There are data on the phenotypic analysis of the wing case pattern of the red soldier beetle on the level of the population and its dependence on the environmental factors [1, 4, 6,8,11].

Insects and small mammals are convenient models for the ecological and biological investigations. Insects are widely-spread, available, show a wide range of variability of the melanistic pattern which makes it possible to analyze the special features of realization of the genetic program of morphogenesis and appearance of phenotypic diversity. Small mammals have close links with various components of ecosystems, a short life cycle, high sensitivity towards environmental changes, a wide range of adaptations and capability of their fast implementation in the new living conditions.

The aim of this work was to study phenetic variability of the animals' populations on the territory of the town of Ishim.

**Materials and methodology of the research**. Material for the research was the following: samples of the firebug (*Pyrrhocoris apterus L.*) gathered in May 2012 in the four districts of Ishim with different anthropogenic burden (railway park, city waste deposit, crossroads of the highways K. Marx street and Artilleriyskaya street, the former garage of agricultural equipment) and a collection of the skulls of the red-backed vole (*Clethrionomys rutilus P.*) and common shrew (Sorex araneus L.) caught in the period from 1996 to 2011 by the method of mass non-selective catch with Gero traps on the territory of the recreational forest "Narodnyi park" in Ishim and in the relict pinewood in the vicinity of the village Sinitsyno of Ishim district, 16 km away from the town (control).

We have analyzed 120 samples of the beetles, 30 in each population. We have described 4 elements of the wing case pattern: the upper and lower black spot, the upper and lower black edging on the red background (fig. 1). Validity of the differences in phenotypic classes within and between the populations was defined using the x2 K. Pearson's criterion [10].

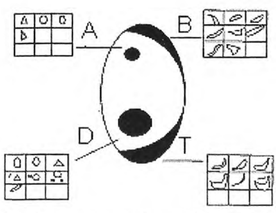


Fig. 1 Variability of the main elements of the wing case pattern of the soldier beetle

We have studied 160 skulls of the red-backed vole (101 from the vicinity of v. Sinitsyno of Ishim district, control; 59 from Narodnyi park). 37 phenes of non-metric skull features of the red-backed vole and 13 phenes of the common shrew have been analyzed. We used the phene catalog developed by A.G. Vasiliev et al. [4]. The phenes showing a significant ($r \geq 0.3$) correlation with the body and skull size and with each other and the phenes which displayed no variability in the studied samples were excluded from the further analysis. Calculation of the median phenetic distances for the red-backed pole was made according to 25 features, and for the common shrew according to 9. Statistical data were processed using Phen and Statan software [3-5].

*Table 1*

**Frequencies of the wing case pattern phenes in the populations of the soldier beetle in Ishim**

| Populations | Frequencies of the wing case pattern phenes | | | | | | | | | | | | | | | | | | | | | | | | | Number of phenes |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Element A | | | | Element B | | | | | | | | Element D | | | | | | | Element T | | | | | | |
| | $A_1$ | $A_2$ | $A_3$ | $A_4$ | $B_1$ | $B_2$ | $B_3$ | $B_4$ | $B_5$ | $B_6$ | $B_7$ | $B_8$ | $D_1$ | $D_2$ | $D_3$ | $D_4$ | $D_5$ | $D_6$ | $D_7$ | $T_1$ | $T_2$ | $T_3$ | $T_4$ | $T_5$ | $T_6$ | |
| 1. Park | 10 | 14 | 6 | | 9 | 7 | 7 | 6 | | | | | 8 | 3 | 13 | 4 | 2 | | | 8 | 6 | 16 | | | | 15 |
| 2. Garage | 12 | 7 | 8 | 3 | 3 | 3 | 5 | 7 | 4 | 4 | 4 | | 7 | 2 | 2 | 6 | 8 | 3 | 2 | 6 | 8 | 7 | 6 | 3 | | 23 |
| 3. Waste deposit | 13 | 2 | 9 | 6 | 4 | 1 | 3 | 5 | 1 | 7 | 5 | 3 | 11 | 8 | 1 | 4 | 1 | 5 | | 2 | 5 | 3 | 4 | 15 | 1 | 24 |
| 4. Crossroads | 24 | 5 | 11 | | 6 | 3 | 8 | 5 | 4 | 2 | | | 18 | 1 | 5 | 4 | 1 | 1 | | 6 | 3 | 1 | 9 | 11 | | 20 |

**Research findings and discussion.** We have singled out 24 phenes of the melanistic wing pattern in the populations of the soldier beetle under study: for element A – 4 phenes; for B – 8; for D – 7 and for T -6 (table 1). The frequency composition of the phenes is different in each population. As for element A, 4 phenes were identified which were met in the populations of the garage (2) and waste deposit (3); and 3 phenes which were registered in the populations of the park (1) and the crossroads (4). Phene $A_1$ dominates in populations 2,3,4 (24%, 26%, 48%); phene $A_2$ (28%) prevails in population 1. 8 phenes were identified in the upper black edging. Only 4 phenes out 8 are present in population 1. As for element B, no prevailing phenes were detected. In the lower black spot the least number of phenes (5) are found in population 1; the biggest number of phenes (7) is found in the population of the waste deposit. Phene $D_1$ dominates in populations 2,3,4. Phene $D_3$ dominates in the park population. 6 phenes were identified in the lower black edging. All the 6 phenes are present in the population of the waste deposit. In the park population only 3 phenes are found. Phene $T_5$ prevails in populations 3 and 4. $T_2$ dominates in population 2 and $T_3$ dominates in population 1.

Results of the research have shown that the biggest variability of the phenetic classes by the 4 elements of the wing case pattern of the soldier beetle is found in the areas of the waste deposit (24 phenes) and garage (23 phenes). 20 phenes were observed in population 4 and 15 phenes were found in the park.

The intra-population analysis on the phenetic frequency has shown proved differences ($P \leq 0.05$) in the park population for phenes A2-A3, D3-D2, D4, D5 and T3-T1, T2. In the garage population the valid differences exist for phenes A1-A4. In the waste deposit population the valid differences are registered for phenes A1-A2, A3-A2, B2-B5, B6, D1-D3, D4, D5 and T5-T1, T2, T3, T4, T6. In the crossroads population the valid differences were identified for phenes A1-A2, A3, D1-D2, D3, D4, D5, D6 and T3-T5, T6.

The inter-population analysis on the phenes frequency has shown similarity of the populations of the waste deposit and garage. Significant differences ($P \leq 0.05$) can be attributed only to phene $T_5$. Between the garage and waste deposit populations significant differences exist only for phenes A3, D2, T3; between the garage and crossroads populations significant differences exist for phenes A1, A3, D1, D5 and T5. The populations of the garage, waste deposit and crossroads differ in the majority of the phenes from the park population. Phenes B8 and T6 have been identified only in the waste deposit population. Phene B7 has been found only in the populations of the waste deposit and garage. Phenes D6 and T5 are specific for the populations of the waste deposit, garage and crossroads.

Thus, the analysis of the melanistic color pattern of the wing case of the soldier beetle in various local populations has shown a wide range of phenotypic variability of this feature. The populations are characterized by different phenetic variability and frequency. The highest phenetic variability level is typical for the populations inhabiting the districts of high anthropogenic burden. The waste deposit population has the highest phenetic variability, especially for phenes D and T. There wastes decay causes emission of methanol and dioxides and changes of the ambient temperature. As a result of dilution of the contaminated substances in rainwater a highly toxic filtrate is formed which accumulates at the bottom of the waste deposit. There is a link between the vestiture melanization and thermal regulation of insects [1-2]. The main factor conditioning high variability of the wing case pattern in the garage and crossroads populations is contamination resulting from motor vehicles [9]. The high phenotypic variability and presence of specific phenes in the areas of the waste deposit, garage and crossroads should be considered as an adaptation strategy of the populations in response to the impact of anthropogenic factors. Possibly the wide variety of melanistic forms is not only of modificational but also mutational nature as the areas of habitation of the populations are full of mutagens of anthropogenic origin. Polymorphism in the populations of the ladybird was earlier registered by N. V. Timofeev-Resovskiy and Ya. Ya. Lusis [8]. In the populations with a high anthropogenic burden we have identified no dominant phenes in the majority of the analyzed elements which may be connected with the change in the selection direction and search of a certain genotypic surrounding.

The phenetic study of the red-backed vole and common shrew has revealed homogeneity of the control and experimental populations of both species regarding the quality composition of phenes and, simultaneously, specificity of each group regarding the correlation of their frequencies. The pair-wise comparisons have demonstrated significant differences between Sinitsyno and Ishim samples of the red-backed vole regarding 6 features (FTmdu – double mental foramen; Fosci – single hole in the occipital bone in the arthrodial fossa; FPm(-) – absence of the alveolar hole; Fasac – presence of an additional alisphenoid hole; FPmb – presence of the alveolar hole); and of the common shrew regarding 1 feature (FMbVI – hole on the surface of the zygapophysis) (tabl. 2, 3). The population of the red-backed vole from the recreational forest differs from the control one in the lower frequencies of all the mentioned phenes, apart from FTmdu – double chin hole. The experimental population of the common shrew is characterized by sharp increase (almost 100-fold) of the frequency of the “rare” for the natural population phene FMbVII. This initializes realization of the rare “diverting” pathways of development and conditions a significant morphogenetic differentiation of the experimental and control populations.

The mean phenetic distances in the whole set of the phenes under consideration between the control and test samples of the red-backed vole and common shrew are statistically valid (tabl. 2, 3).

*Table 2*

**Comparison of different samples of the red-backed vole regarding the frequencies of the phenes of non-metric skull features**

| Feature | Frequency % | | Comparison | |
|---|---|---|---|---|
| | control | test | MMD | $\chi^2$ |
| Fpodu | 5,08 | 3,57 | 0, 0099 | 0,29 |
| Ffran II | 48,11 | 58,62 | 0,0299 | 3,14 |
| Ffran(-) | 7,98 | 6,03 | -0,0091 | 0,34 |

| | | | | |
|---|---|---|---|---|
| Ffrdu | 53,26 | 58,41 | -0,0037 | 0,74 |
| FTm(-) | 0,56 | 0,95 | -0,012 | 0,20 |
| FTmdu | 1,67 | 6,67 | 4,5* | -0,01 |
| MeTm(-) | 17,88 | 16,98 | -0,015 | 0,03 |
| Fsqor | 25,54 | 33,65 | 0,017 | 2,12 |
| Fosci | 18,3 | 5,15 | 0,153 | 10,1** |
| Fcnsu | 50,32 | 48,39 | -0,016 | 0,09 |
| Fhgsi | 45,16 | 26,6 | 0,131 | 8,73** |
| FPm(-) | 17,77 | 8,62 | 0,057 | 5,19* |
| Fmxdu | 56,06 | 59,82 | -0,008 | 0,40 |
| Fpmme | 95,96 | 98,25 | 0,001 | 1,08 |
| FePl | 2,12 | 9,26 | 0,073 | 3,51 |
| MgPl | 8,85 | 6,67 | 0,014 | 0,29 |
| LlnFOv | 10,98 | 12,12 | -0,015 | 0,098 |
| LtvFOv | 5,49 | 9,09 | 0,004 | 1,25 |
| Fasac | 47,24 | 29,59 | 0,114 | 8,01** |
| Fmtdu | 14,65 | 18,97 | 0,0002 | 1,01 |
| Fmtla | 1,52 | 4,24 | 0,015 | 2,08 |
| Fmtan | 58,42 | 48,31 | 0,027 | 3,05 |
| Fmtlg | 40,59 | 39,83 | -0,013 | 0,016 |
| FPmb | 35,15 | 24,58 | 0,039 | 3,9* |
| MMD | 0,0247* | | | |
| MSD | 0,00449 | | | |

Note: * — variations in the frequency of a phene are valid at $p<0.05$; ** — at p,0.01; MMD — mean phenetic distance on the set of 25 features; MSD – aggregate standard deviations; differences are considered valid as a rule at $p<0.05$ if MMD>2MSD.

*Table 3*

**Comparison of different samples of the common shrew regarding the frequencies of the phenes of non-metric skull features**

| Feature | Frequency % | | Comparison | |
|---|---|---|---|---|
| | control | test | MMD | $\chi^2$ |
| 1. FMt | 92,0 | 95,0 | -0,0029 | 0,79 |
| 2. FMbII | 9,0 | 6,0 | -0,007 | 0,52 |
| 3. FMbIII | 3,0 | 6,0 | 0,0007 | 1,05 |
| 4. FMbV | 9,0 | 13,0 | 0,0013 | 1,09 |
| 5. FMbVII | 1,0 | 99,0 | 7,27 | 528,23*** |
| 6. FMbVIII | 83,0 | 87,0 | -0,025 | |
| 7. FTman | 57,0 | 57,0 | 0,068 | 3,41 |
| 8. FPan | 55,0 | 39,0 | 0,255 | 9,67 |
| 9. FPpo | 67,0 | 40,0 | -0,023 | 0,042 |
| MMD | 0,0577*** | | | |
| MSD | 0,0074 | | | |

As for the red-backed vole the aggregate phenetic distance between the Ishim and Sinitsyno population groups located 16 km away from each other correspond to the differentiation level between neighboring populations isolated by a 30-40 km distance on the continuous habitation area (MMD = 0.011-0.029). As for the common shrew the phenetic differentiation approaches the level defined by A.G.Vasiliev for neighboring populations isolated by 30-40 km of the landscape-ecological barrier (MMD= 0.060-0.075) [3-4].

The results which we obtained correspond to the scientific data on an unexpectedly high level of differentiation of the local populations of small mammals in cities as compared to similar populations outside the urban zones [4]. This fact proves that urbanization is a real integral factor speeding epigenetic differentiation of populations.

**Conclusions**:

1. In the areas with a high anthropogenic burden we have registered an increased variability of the phenotypic color pattern of the wing cases of the soldier beetle and emergence of rare phenes, which indicates a rather adaptive than neutral reaction of the population towards an unfavorable environment.

2. In the populations of the soldier beetle we have identified phenes which can serve as specific markers (B8; T6; B7; D6 and T5) of anthropogenic pollution.

3. The populations of small mammals in the urban territories are characterized by a specific phenetic structure.

4. The urban populations of the red-backed vole and common shrew differ from the natural ones in significant phenogenetic differentiation exceeding the level corresponding to the degree of their spatial-geographic isolation. This fact lets us consider urbanization a factor increasing epigenetic differentiation of animal populations.

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