

Technology for Restoring and Maintaining Sustainability of Populations: Practical and Theoretical Results of Genourbanology

V. M. Makeeva^{1,*}, A. V. Smurov¹, D. V. Politov², M. M. Belokon², Y. S. Belokon², E. G. Suslova³, and A. A. Kalinin⁴

¹Museum of Earth Sciences, Lomonosov Moscow State University, Leninskiye Gory 1, Moscow, 119991, Russia

²Vavilov Institute of General Genetics, Russian Academy of Sciences (RAS IOGen), Gubkina st., ap. 4, Moscow, 119991, Russia

³Faculty of Geography, Lomonosov Moscow State University, Leninskiye Gory 1; Moscow, 119991, Russia

⁴Nonprofit Nature Conservation Foundation "Verkhovie," Agrohnikov st., ap. 6, settlement Novoivanovskoe, Odintsovsky region, Moscow oblast, 143026, Russia

Abstract: Genourbanology (the synthesis of system ecology and population genetics) is a new scientific and practical school of thought, founded and developed by the authors. The task of genourbanology consists in studying the genetic parameters and laws of stability conservation of ecosystems and ecosystem restoration in anthropogenic and, in particular, urbanized landscapes. An ecological genetic technology for restoring and maintaining sustainability of populations viability, including the method for assessing the state and duration of existence of populations as well as the enrichment and control of the population gene pool in urban ecosystems was developed in the framework of genourbanology. The results of quantitative assessment of the state of the gene pool of 40 populations are described and the recommendations for restoring its viability are given using the following model species of animals and plants inhabiting the fragmented landscape of Moscow and Moscow region: 20 populations of the terrestrial bush snail (*Bradybaena fruticum* (Mull.)), 16 populations of brown frogs *Rana temporaria* L. and *Rana arvalis* Nills., and 4 populations of Norway spruce *Picea abies* (L.) Karst.). A sharp decline in the genetic diversity (up to 70%) in the urban isolated compared to the natural populations was found. Comparative analysis of the gene pool of natural populations and planted spruce stands revealed a correlation between the state of the gene pool of spruce populations and the degree of their lesion with the bark beetle (*Ips typographus* (L.)).

Keywords: Gene pool, genourbanology, isozymes, population, urban landscape

INTRODUCTION

Biodiversity conservation is proclaimed as one of the essential conditions for sustainable human development [1]. The main prerequisite for the conservation of biodiversity is the conservation of the genetic diversity of populations, which ensures their adaptation to the environment [2]. Under conditions of global urbanization that swept over 60% of the planet, the development of effective approaches to conserving the biodiversity of anthropogenic (human-dominated) ecosystems is one of the key objectives of modern ecology.

There is a fundamental difference in the approaches to the conservation of natural and urban landscapes.

In *natural landscapes*, the main method of biodiversity conservation that ensures the sustainability of ecosystems is

the territorial conservation based on the self-recovery of ecosystems [3].

However, to conserve the biodiversity in *urban landscapes* and in a number of natural landscapes, it is necessary to take into account the state of the population-genetic level of the organization of ecosystems and, if required, to conserve and restore the quality of the gene pool of the conserved populations [4]. These provisions were substantiated by us in the framework of genourbanology, a recently developed new scientific and practical direction [4–6]. The purpose of genourbanology is to understand the genetic parameters and consistent patterns of conservation of sustainability and restoration of ecosystems of anthropogenic and especially urban landscapes.

The urgent need to develop a new ecological genetic approach to biodiversity conservation in urban landscapes arose in connection with the creation at the beginning of the 21st century of protected areas in many cities of Russia and Europe. In Moscow, the network of specially protected areas, in which a thorough territorial conservation is carried out and the habitats and community structure are preserved,

*Address correspondence to this author at the Museum of Earth Sciences, Lomonosov Moscow State University, Leninskiye Gory 1, Moscow, 119991, Russia; Tel: +7(495)939-1415; Fax: +7(495)939-1415; E-mail: vmmakeeva@yandex.ru

covers 16% of the city. Despite this, the proportion of species of vertebrate animals at the brink of extinction ranges from 40 to 70%, for both urban specially protected areas in Moscow and in Europe [7].

A distinctive feature of urban specially protected areas is that small areas are in the form of separate isolated islands in a highly urbanized landscape, and the conservation of biodiversity in them is combined with the active use of these areas for the recreational purposes. The fact of habitation of animals in small isolates in newly created specially protected urban areas was not taken into account, and the theory of the scientific organization of biodiversity conservation in them was still based on the concept of territorial conservation [8].

This concept does not take into account the action of well-known theoretical laws of population genetics in isolates, which cause rapid changes in the structural and functional properties of the gene pool due to genetic drift and inbreeding [9–11], reducing the viability and leading to inevitable extinction of populations.

For the genetic aspects of nature conservation, it is important to know that a reduction in the gene pool diversity (decrease in heterozygosity) is associated with practically important properties, such as decrease in the viability, fertility, and growth rate [2]. The availability of information about changes in the quantitative parameters of the gene pool makes it possible to outline the strategy for its stabilization and restoration. The concept of optimal evolutionary occurred genetic diversity as an important condition of successful existence of populations in a normally fluctuating environment, developed by Academician Yuri Petrovich Altuhov, is the basis for the development of ecological and genetic aspects of biodiversity conservation in urban landscapes.

The rationale for distinguishing a new scientific and practical direction, genourbanology, was a series of papers [4, 12, 13] taking into account the evaluation of the state of the gene pool of the populations studied [5, 14, 15], the prediction of the duration of their existence, and experimental work on the improvement of the gene pool of populations [4, 16, 17], as well as the development of ecological and genetic approach to the wildlife conservation in urban landscapes [4; 18–20]. Below are summarized the theoretical and practical results of a 30-year ecological genetic monitoring conducted for model plant and animal species.

The study of the state of the gene pool of urban populations of animals and the substantiation of genourbanology were performed in accordance with the state program "Restoration of Biodiversity in Moscow."

In the framework of genourbanology, we have developed and tested an ecological genetic approach combining the theoretical and practical aspects of biodiversity conservation (methodology, concept, strategy, and the technology for maintaining the sustainability of populations, which was tested in the system of urban specially protected areas in Moscow [4, 17, 18]).

MATERIALS AND METHODS

The gene pool of populations of species of living organisms inhabiting urban and reference natural ecosystems is the subject of genourbanology. To study the state of the gene pool of urban populations of animals, a system of urban ecological genetic monitoring consisting of 36 populations of model species, of which 20 are urban and 16 are natural, which were collected in twelve Moscow parks and five localities of northwestern Moscow region, was created for the first time in Russia and in the world. (For detailed maps of the study area see our earlier publications [5, 6, 15]).

Animals

The bush snail *Bradybaena fruticum* (Müll.), Mollusca (since 1975) and two species of brown frogs—the moor frog *Rana arvalis* Nills. and the common frog *Rana temporaria* L. (since 2003) - are the model objects for the study of the gene pool of natural and urban populations of animals. The volume of the material studied is more than 30000 snail shells and more than 9000 electrophoretograms obtained by the electrophoresis of proteins (1975–2005). The populations of snails (approximately 3000 specimens) were analyzed for 13 polymorphic isozyme loci (2002–2003), and 554 frogs were analyzed for 7 polymorphic loci.

Plants

In 2012, the state of the gene pool of plant populations was studied using the Norway spruce (*Picea abies* (L.) Karst.) as an example [21]. In total, we examined four populations of spruce from two districts of Moscow region (Ramenki and Odintsovo). In each area, we studied one quasi primary (conditionally natural) population as well as a forest plantation (Ramenki district) and a short-term population (Odintsovo district). In Odintsovo district (urban settlement Zvenigorod), we examined conditionally native spruce–pine forests 100–170 years of age with pine, oak, and lime trees, overgrown with small shrubs, ferns, shamrock, and broad grass [22]. At a distance of 760 m from the native (reference) stand, short-term spruce plantations and subnemoral aspen–birch–spruce forests 70–90 years of age overgrown with shamrock, ferns, and weaselsnout were examined.

In Ramenki district (urban settlement Kratovo), we examined spruce–pine subnemoral forests interspersed with pine–fir and spruce forest fragments aged 70–120 years. At a distance of 5 km from the reference stand, transformed spruce–pine plantations older than 90 years were examined. We studied 37 specimens of *Picea abies*, each taken from one tree, for 24 polymorphic isozyme loci in each population [21].

Method for Assessing the State and Duration of Existence of Populations in Urban Ecosystems

The method for assessing the gene pool consists in comparing the genetic parameters of variation of urban populations with large natural reference populations [5, 15, 23]. It is the main genourbanology method, which makes it

possible to determine the parameters of stability of the genetic basis of anthropogenic ecosystems [4]. The genetic diversity of urban populations was estimated by the sum of quantitative indices, such as the proportion of polymorphic loci, the mean and effective number of alleles per locus, and the observed and expected heterozygosity [6, 24]. To assess the Hardy – Weinberg equilibrium we used Chi-square tests [24].

The duration of existence of populations (prediction) was calculated using the formula proposed by M.E. Sulei [25]:

$$t = 1.5 N_e,$$

where t is the number of generations and N_e is the effective population size.

METHOD OF ISOZYME ANALYSIS OF ANIMAL MUSCLE SAMPLES

Electrophoretic separation of isozymes was performed in 13% starch or 7.5% polyacrylamide gels [15, 26]. It was assumed that the zones of activity of enzymes are controlled by individual gene loci; they were designated in accordance with the EC [27].

The standard genetic distances [28] were used to determine genetic similarity between the populations studied. The results were statistically processed using the standard GenAIEx V5 [29-30] and POPGENE [31] software.

RESULTS AND DISCUSSION

The Study of Animal Populations

Long-term ecological genetic monitoring revealed a global trend—a reduction in the genetic diversity of animal populations (as exemplified by model species) in the isolates of fragmented landscape of Moscow and Moscow region [5, 13, 32]: the gene pool diversity of 77% populations in the city is reduced at least by 50%.

The increase in homozygosity found in 67% populations of the bush snail reaches 28%; in 25% of these populations, homozygosity is increased by more than 25%. However, there is experimental evidence that the viability of populations decreases by 25% when their homozygosity increases by only 10% [33].

Our study showed that the reduction in the gene pool diversity in urban populations is caused by the genetic drift and inbreeding, which accompanies it. These negative processes are the key processes in populations as a result of anthropogenic fragmentation of landscape.

The prediction of the duration of existence of the urban populations of three model species confirmed the degradation of the gene pool of urban populations occurred as a result of anthropogenic isolation [4, 6, 16]: in the next 100 years, over 56% of populations may disappear, 28% of which may disappear within 25 years, and approximately 84% of populations may disappear within 160–200 years. Only 16% of populations have a chance of existence for 500 years. This prediction is real today at the currently existing standard approach to the protection of animals (i.e., territorial conservation).

The determination of the degree of deviation of the gene pool parameters of anthropogenic ecosystems from those of the reference ecosystems makes it possible to develop measures aimed at restoring the gene pool quality and, therefore, the viability of populations and conservation of biodiversity in urban areas. If the diversity of the gene pool of urban populations is reduced by 10–20%, its quality can be estimated as stable, a decrease by 20–60% indicates that its state is critical, and a decrease of diversity to a level of 60% and more can be regarded as evidence of its catastrophic state. Apparently, the last two cases require human intervention and artificial restoration of the gene pool quality and, hence, the population viability.

Results of the experiment on the restoration (enrichment) of the gene pool of four populations of shrub snails inhabiting specially protected natural areas of Moscow confirmed the effectiveness of using the developed technology for restoring and maintaining the viability of populations in urban landscapes. The control of the shift in allele frequencies on the basis of shell banding was performed in juvenile snails of the next generation; in all cases, a significant shift in the frequency of the allele responsible for shell banding, which was used as a marker for studying the changes of the gene pool, was observed [4, 16, 18].

The restoration (health improvement) of the gene pool of the bush snail was performed in accordance with the guidelines developed for improving the gene pool health, most of which are universal and are described below.

It should be noted that, once started, negative genetic processes in isolates (genetic drift and inbreeding) continue to the end (i.e., they are irreversible) [10]. Therefore, in all urban areas of the Earth, the quality of the gene pool undergoes irreversible degradation, which manifests itself in the reduction of its genetic diversity. This postulate is fundamental in developing the modern strategies for the conservation of biodiversity, because it allows researchers to understand that this irreversible process can be stopped only artificially, i.e., with the aid of humans.

The Study of Plant Populations

It was found that the examined native populations of Norway spruce from two districts of Moscow region do not differ by the set of allele frequencies. The identified genetic indices are close to those characteristic for the natural populations of the Central Region of the East European forest zone [34, 35, 36]. The indices characterize the optimum of the evolutionary formed genetic diversity as the key prerequisite of successful existence of a population in a normally fluctuating environment [2].

The stands differed in the degree of damage by European spruce bark beetle *Ips typographus* (L.) which was high (90%) in the plantation Kratovo (90%), low in a short-term stand of Zvenigorod and almost absent in corresponding virtually native stands. It is noteworthy that averaged over loci Wright's fixation index F (intrapopulation inbreeding coefficient) was positive and twice as high in a plantation (0.087) and a derived population (0.100) as compared to the corresponding native stands (0.040 and 0.050), (see Table 1) Chi-square test showed significant deviation from Hardy –

Table 1. By-locus values of observed (H_O) and expected (H_E) heterozygosity and the inbreeding coefficient (F).

| Loci | Samples | | | | | | | | | | | |
|---------------|--|-----------|-----------|--|-----------|----------|--|-----------|-----------|---|-----------|-----------|
| | Kratovo Forest Plantation Parasitized | | | Kratovo Natural Stand Non Parasitized | | | Zvenigorod Short-terms Forest Plantation Parasitized | | | Zvenigorod Natural Stand Non Parasitized | | |
| | H_O | H_E | F | H_O | H_E | F | H_O | H_E | F | H_O | H_E | F |
| <i>Fdh</i> | 0.216 | 0.234 | 0.075 | 0.216 | 0.234 | 0.075 | 0.189 | 0.214 | 0.115 | 0.297 | 0.323 | 0.080 |
| <i>Fe-2</i> | 0.167 | 0.153 | -0.091 | 0.216 | 0.217 | 0.002 | 0.216 | 0.243 | 0.108 | 0.135 | 0.127 | -0.060 |
| <i>Gdh</i> | 0.432 | 0.394 | -0.096 | 0.324 | 0.368 | 0.119 | 0.270 | 0.272 | 0.005 | 0.378 | 0.418 | 0.094 |
| <i>Got-1</i> | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - |
| <i>Got-2</i> | 0.054 | 0.053 | -0.028 | 0.000 | 0.000 | - | 0.027 | 0.027 | -0.014 | 0.027 | 0.027 | -0.014 |
| <i>Got-3</i> | 0.568 | 0.502 | -0.130 | 0.595 | 0.497 | -0.197 | 0.297 | 0.499 | 0.404 * | 0.405 | 0.500 | 0.189 |
| <i>Idh-1</i> | 0.135 | 0.290 | 0.533 ** | 0.135 | 0.126 | -0.072 | 0.081 | 0.126 | 0.357 | 0.000 | 0.000 | - |
| <i>Idh-2</i> | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.054 | 0.053 | -0.021 | 0.000 | 0.000 | - |
| <i>Lap-1</i> | 0.243 | 0.304 | 0.200 | 0.216 | 0.240 | 0.098 | 0.162 | 0.316 | 0.486 ** | 0.135 | 0.220 | 0.386 ** |
| <i>Lap-2</i> | 0.405 | 0.504 | 0.196 | 0.378 | 0.449 | 0.157 | 0.189 | 0.294 | 0.357 | 0.351 | 0.420 | 0.164 |
| <i>Mdh-1</i> | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - |
| <i>Mdh-2</i> | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - |
| <i>Mdh-3</i> | 0.162 | 0.244 | 0.334 ** | 0.324 | 0.441 | 0.265 | 0.189 | 0.177 | -0.068 | 0.270 | 0.246 | -0.100 |
| <i>Pepca</i> | 0.135 | 0.171 | 0.211 | 0.081 | 0.078 | -0.042 | 0.216 | 0.193 | -0.121 | 0.162 | 0.149 | -0.088 |
| <i>6Pgd-2</i> | 0.389 | 0.498 | 0.220 | 0.556 | 0.498 | -0.115 | 0.459 | 0.513 | 0.105 | 0.514 | 0.526 | 0.024 |
| <i>6Pgd-3</i> | 0.278 | 0.422 | 0.341 *** | 0.111 | 0.198 | 0.438 ** | 0.486 | 0.456 | -0.067 | 0.270 | 0.467 | 0.422 *** |
| <i>Pgi-1</i> | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - |
| <i>Pgi-2</i> | 0.757 | 0.629 | -0.203 ** | 0.722 | 0.629 | -0.148 | 0.703 | 0.654 | -0.074 | 0.757 | 0.622 | -0.217 |
| <i>Pgm-1</i> | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - |
| <i>Pgm-2</i> | 0.270 | 0.285 | 0.051 | 0.351 | 0.304 | -0.155 | 0.297 | 0.266 | -0.120 | 0.243 | 0.264 | 0.079 |
| <i>Skdh-1</i> | 0.054 | 0.053 | -0.028 | 0.054 | 0.053 | -0.028 | 0.054 | 0.102 | 0.471 ** | 0.081 | 0.078 | -0.042 |
| <i>Skdh-2</i> | 0.027 | 0.027 | -0.014 | 0.081 | 0.127 | 0.364 | 0.108 | 0.104 | -0.042 | 0.108 | 0.104 | -0.042 |
| <i>Sod-1</i> | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - | 0.000 | 0.000 | - |
| <i>Sod-2</i> | 0.162 | 0.149 | -0.088 | 0.216 | 0.193 | -0.121 | 0.135 | 0.126 | -0.072 | 0.135 | 0.126 | -0.072 |
| Mean | 0.186 | 0.205 | 0.087 | 0.191 | 0.194 | 0.040 | 0.172 | 0.193 | 0.100 | 0.178 | 0.192 | 0.050 |
| (± SEM.) | (±0.041) | (± 0.041) | (±0.041) | (±0.043) | (± 0.040) | (±0.039) | (± 0.037) | (± 0.038) | (± 0.044) | (± 0.040) | (± 0.041) | (±0.035) |

Levels of probability: * P < 0.05, ** P < 0.01, *** P < 0.001
 - F cannot be calculated as the locus's heterozygosity is zero.

Weinberg proportions in a plantation Kratovo (4 cases, 3 of them due to heterozygote deficiency), short-terms stand (5 cases all related to heterozygote deficiency), as compared to 2 significant cases of deviations coupled with positive F_{IS} values in each of two native stands. Various loci were involved in deviation from equilibrium state and by only locus *Idh-1* showed significant inbreeding in both non-native stands indirectly indicating its putative adaptive value.

Chi-square test for heterogeneity of allelic frequencies have shown significant differentiation between damaged plantation and undamaged native stand in Ramensky district

by several loci (*Fe-2*, *Idh-1*, *Mdh-3*) and for all loci in total. In Odintsovsky district, slightly damaged short-terms stand also differed from native stand by some loci (*Gdh*, *Idh-1*).

The results of the study showed that the conditionally natural and the short terms stands are more resistant to the damage by the bark beetle than the planted forests. However, other than genetic factors such as multi-species and multi-age structure of native stands definitely contribute a large portion of sustainability of these forests. The altered genetic composition of secondary forests and especially in its extreme form of artificial plantation may have negative

effects on viability of trees and their susceptibility to pests and pathogens.

Thus, the preliminary results of the comparative analysis of natural populations of plants and planted forests using the Norway spruce *Picea abies* (L.) Karst. from the Moscow region as an example, which were obtained as a result of a small number of populations, allowed us to identify the relationship between the state of the gene pool of spruce and the degree of its damage by the bark beetle (*Ips typographus* (L.) and demonstrated the necessity to take into account the environmental structure of populations [37].

The formation of viable forest cultures and restoration of plant populations in urban landscapes disturbed by human activities should be performed taking into account the state of the genotypes of the plant material used for these purposes [38]. In this connection, the study devoted to the development of genetic markers for certification of forest plantations is of particular importance [39]. The authors confirmed the effectiveness and feasibility of using microsatellite loci as molecular markers for genetic certification of seed-production plantations and identification of seed lots used in reforestation.

The results of the practical study of the state of gene pools of populations of plants and animals were used to develop a strategy for the conservation of biodiversity of anthropogenic ecosystems, which will be discussed below.

Theoretical Results of Genourbanology

The methodology of genourbanology includes the ecological and genetic principles, the ecological and genetic concept, and the strategy and technology of restoring and maintaining the viability of populations in urban landscapes.

The systemic approach to the organization of biocos and living systems of the Earth is the basis for the substantiation of the methodology of genourbanology. The idea of the systemic organization of the hierarchical structure of the biosphere [40, 41] allows regarding a population as an elementary structural living unit maintaining the homeostasis of systems of higher hierarchical levels of the biosphere (ecosystems and biogeosystems). The identification of the relationship (forward and backward) between all levels of the biosphere organization allows defining the main task of genourbanology as the determination of genetic parameters and mechanisms for maintaining the homeostasis of anthropogenic ecosystems.

It should also be noted that the destruction of the hierarchy of the population structure of species in the urban landscape of Moscow and Moscow region was revealed using genetic distances [28] (for detailed tables see [5, 6, 15]). It also justifies the priority of the population level of organization of urban ecosystems in organizing the conservation of biodiversity in urban landscapes. In addition, these studies apparently have contributed to the development of the theory of species and speciation under modern conditions (i.e., urbanization).

The methodology of genourbanology is based on the fundamental nature-conservation principles developed for

the ecosystem and population-genetic levels of conservation [2, 42]: the principle of the universal connection (forward and backward) of hierarchical levels and the interaction of all natural elements, the principle of diversity of structural elements, and the principle of potential usefulness of each component.

The ecological genetic principles for the conservation of biodiversity developed by us take into account the characteristics of existence of populations in anthropogenically disturbed ecosystems [4, 43]. Unlike the territorial approach, they require the conservation of a minimum number of populations that could provide a minimum loss of genetic diversity. A decline of abundance below its minimum level makes it necessary to maintain the gene pool quality solely by artificial means (creation of artificial gene flows).

Ecological Genetic Concept of Conservation

The developed concept of biodiversity conservation is based on analysis of data obtained as a result of

(1) comparative study of the structural and functional state of the gene pool (as exemplified by model species) under conditions of natural and urban landscapes [5, 14, 15, 44, 45] and

(2) study of consistent patterns (factors and mechanisms) of changes in the diversity of the gene pool of populations under conditions of anthropogenic fragmentation of landscape [5, 14, 15, 23, 46, 47].

The ecological genetic concept of biodiversity conservation (including animals) is to recognize the possibility of cessation of inevitable extinction of populations and the possibility of restoration of their adaptive capacity, viability, and long-term sustainable existence. The long-term viability of populations can be restored only by switching from the purely territorial conservation and noninterference to active restoration of the gene pool of populations. The ecological genetic concept does not rule out the territorial conservation, which is the main type of conservation at the ecosystem and biogeocenotic level.

It should be noted that the substantiation of the concept of biodiversity conservation in anthropogenic ecosystems requires the recognition of the new modern complex ecological genetic paradigm, which takes into account the role of genetic causes in the reduction in the Earth's biodiversity [4, 18, 20].

The development of the population genetic level of conservation is important because in future the humankind will live almost exclusively in anthropogenic landscapes.

Genourbanology ensures the conservation of biodiversity of urban landscapes taking into account the population genetic level of organization of ecosystems, which ensures a new, deeper level of development of ideas, methods, and principles of conservation of the nature, biodiversity, and anthropogenic ecosystems as a whole and includes [4].

(1) development of methods and criteria for assessing the state of the gene pool of populations and the necessity of its improvement [4–6, 14–16, 46, 47];

(2) development of genetic methods for restoring and maintaining long-term viability of populations of endangered animal species inhabiting specially protected urban natural areas [4, 16, 18];

(3) development of the ecological genetic aspect of the biodiversity conservation strategy, including the conservation of the animals inhabiting degrading urban protected ecosystems and the restoration of sustainability of ecosystems as a whole [44]; and

(4) development of a new system of nature-conservation ecological genetic measures aimed at restoring and maintaining the long-term viability of populations of endangered species, including those listed in the Red Data Book of the city [4, 7, 18, 19].

The results of our studies on assessing the state of the gene pool and the scientifically substantiated prediction of the duration of existence of urban populations of animals served as a basis for changing the current strategy for conservation of animals inhabiting the specially protected areas in Moscow.

Strategy for Biodiversity Conservation in Anthropogenic Ecosystems

The prerequisites for the realization of the ecological genetic aspect of biodiversity conservation are the identification of a strong vulnerability of the gene pool and the exceeding of the ultimate strength of the gene pool: approximately 80% of urban populations (primarily animal model species) have lost more than half of their genetic diversity.

The goal of the developed environmental genetic strategy is the creation and implementation of genetic technologies, i.e., the mechanisms of recovery and long-term maintenance of viability of natural populations of species in anthropogenic ecosystems, including the endangered species.

The main strategic task is the restoration and maintenance of the quality (diversity) of the gene pool of protected natural populations as a basis for the restoration and conservation of natural degrading urban ecosystems.

The strategy of conservation of the biodiversity of anthropogenic ecosystems is aimed at transition from the purely passive territorial conservation and noninterference to active restoration of the gene pool of populations, i.e., the restoration of their viability and sustainable existence. Certainly, the territorial conservation is required, but it cannot be the sole and sufficient measure for biodiversity conservation in urban landscapes.

The strategy aims to prioritize the population genetic principle of conservation of species diversity (primarily animal) in specially protected urban areas and the methods for its conservation in natural habitats.

On the basis of the results of the practical study of the gene pool of populations of plants and animals, a general

strategy for protecting the biodiversity of anthropogenic ecosystems has been developed [4, 44], the main features of which are the active management of the quality of the gene pool of protected populations and the maintenance of the viability of populations with the aid of modern technologies along with the territorial conservation.

Of course, there are certain differences in the organization of the gene pool management and in specific technologies used for restoration in different groups of organisms.

Our previous research showed that the strategy for the conservation of animals of anthropogenic ecosystems on the Earth [4, 20, 44] should be aimed at switching from the purely passive territorial conservation and noninterference to the active restoration and sustainable conservation of the gene pool of populations.

Modern forest management strategy should be focused on the formation and maintenance of the "optimal" genetic diversity of populations, characteristic of a given natural zone, along with the maintenance of the maximum species richness, age difference, and other characteristics of natural forests, which determine their spatial conjugation [21].

The results of our studies showed that the priority measures aimed at conserving species diversity (including rare and endangered species) should include the development and application of genetic technologies for restoration and maintenance of a long-term (sustainable) existence of natural populations inhabiting protected areas of anthropogenic landscapes [4, 16].

Principles and Techniques for Long-term Conservation and Maintenance of Viability of Natural Populations of Anthropogenic Ecosystems. Areas of Application

The main principle of sustainable use of natural populations of (unexploited) animal and plant species inhabiting the protected ecosystems of anthropogenic landscapes is the principle of the necessity of diversity of structural elements at the population-genetic level of organization of anthropogenic ecosystems.

The realization of this principle is associated with the maintenance of the genetic parameters of sustainability of populations inhabiting anthropogenic ecosystems, which are also the parameters of sustainability of ecosystems.

This principle cannot be implemented in urban areas with the aid of the territorial conservation.

The main condition for the implementation of the new environmental genetic strategy is the development and introduction of new genetic technologies for the conservation and sustainable use of genetic resources in anthropogenic ecosystems.

Technology for restoring and maintaining the viability of populations of urban landscapes:

The technology involves several successive stages including the determination, restoration, maintenance, and control of the genetic parameters of population sustainability [6, 18].

To determine the genetic parameters of the state of the gene pool of populations, our method for assessing the state and duration of existence of populations should be used (see above).

To restore the parameters of sustainability of populations (i.e., the optimal state of the gene pool characteristic for a given zonal ecosystem), it is necessary to organize an artificial gene flow from nature or a zooculture. The developed and tested practical recommendations are given below.

To maintain certain genetic parameters ensuring the viability of populations, it is necessary to monitor the state of their gene pool, which should be exercised by means of ecological genetic monitoring.

Areas of application: the use of the technology in the organization of scientific management of urban specially protected areas, implementing biodiversity conservation during intensive use of their areas for the purpose of recreation as well as for the conservation and restoration of biodiversity in any anthropogenic areas experiencing anthropogenic impact of varying severity.

Key directions of action in the practical and economic sphere of the conservation of biodiversity of anthropogenic ecosystems are the introduction of relevant principles and technologies from the conservation of the gene pool of populations to the organization of scientific management of urban specially protected areas. The genetic principles and technologies should be taken into account in planning the measures for sustainable use of natural biological resources and development of new areas.

The necessity to introduce new technologies is determined by the scale and severity of anthropogenic transformation of the gene pool of populations.

Objects of conservation and restoration in anthropogenic ecosystems are the widespread species that are endangered or whose abundance declines as a result of anthropogenic impact (i.e., the species that were not rare earlier but have become rare as a result of impact of anthropogenic limiting factors).

In addition, it is necessary to protect and restore the naturally rare species, potentially vulnerable due to their biological characteristics and less able to withstand anthropogenic impact (narrow-range, endemic, highly specialized, and stenobiotic species).

Guidelines for restoration of genetic parameters of population sustainability (restoration of their genetic diversity):

(1) The set of populations of the species that are subject to restoration in the given ecosystem should be distinguished. It is necessary to analyze the populations of not only endangered but also the key species of ecosystems;

(2) The state of the gene pool should be assessed for the largest possible number of enzymes and loci (at least 5–6 enzymes and 10–12 loci);

(3) A prerequisite for enrichment is the genetic analysis of donor populations (i.e., large natural populations, preferably from the same region, living in similar climatic

conditions). The results of our studies showed that the gene pool of donor populations (approximately 23%) inhabiting the anthropogenic landscapes of Moscow region may be highly depleted;

(4) It is necessary to take bear in mind that opposite changes in the allele frequencies of the same locus may occur in different populations due to fixation of alleles during genetic drift;

(5) For enrichment, it is advisable to use several donor populations with opposite trends in fixing alleles of the same locus;

(6) In the future, it is necessary to monitor the state of the gene pool of the enriched populations.

These recommendations were tested in the specially protected areas of Moscow (2003–2005) under the program "Restoration of Biodiversity in Moscow." Currently, they are used in the restoration of biodiversity in Moscow and other major Russian cities, including St. Petersburg.

Taking practical measures aimed at enriching the populations of endangered species (primarily animals) will make it possible to increase heterozygosity and, therefore, the viability of populations. This will help to restore all relationships (nutritional, energy, informational, etc.) in ecosystems and, hence, the sustainability of ecosystems as a whole.

CONCLUSION

At the present stage of drastic transformation of natural zonal ecosystems, which has swept over half of the planet, the priority level of biodiversity conservation, along with the territorial one, should be the population genetic level of organization of ecosystems. The ecological genetic approach to the conservation of biodiversity, which is the prerogative of the new scientific and practical direction, genourbanology, should ensure the sustainable use of the genetic resource of anthropogenic ecosystems, which is a prerequisite for sustainable conservation of biodiversity and human civilization.

The technology developed to restore and maintain the viability of populations in urban landscapes makes it possible to stop the irreversible genetic processes in populations, which decrease the adaptive capacities of populations and lead to their inevitable extinction. It is focused on the restoration of the gene pool of those populations whose minimum abundance is insufficient to ensure the optimal level of genetic diversity characteristic of specific zonal ecosystems.

The restoration of the genetic parameters of ecosystems with the aid of genetic technologies makes it possible to interrupt the destructive chain reaction that is triggered by the stochastic loss of rare alleles, continues by the loss of rare species, and is ended with the collapse of the existing evolutionary formed relationships and the degradation of ecosystems in urban landscapes.

Thus, genourbanology is the real basis for the conservation and restoration of biological resources not only in urban landscapes but also in any anthropogenically modified communities and ecosystems. The principles,

approaches, and technologies developed in the framework of this direction should be taken into account when planning the transformation of landscapes and the development and sustainable use of biological resources.

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

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