

A study of the effect of habitat fragmentation on the population status of *Iris pumila* L. in Ukraine

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Abstract

The goal of the study was to investigate the effect of isolation on populations of *Iris pumila* L., a typical Ukrainian steppe xerophyte which habitat has been split into a multitude of small fragments. Most of the studied populations clearly demonstrate prevalence of adult flowering plants. Seedlings are rare, which can be explained by drought during major parts of the vegetative seasons, the substantial matting of local soil, and human impacts, such as burning. Population success appears to depend on the size of the steppe fragment and the total human impact. In this preliminary genetic study of a relatively large *Iris pumila* populations no signs of gene pool depletion were detected. Further research will hopefully reveal whether the genetic indices that have been estimated are also useful tools for other populations of the species – fragments of a previously continuous habitat. Meanwhile, due to the increasingly endangered status of the habitat and the practical absence of any population status monitoring, it makes sense to include this species into the Ukrainian Red List.

Keywords: *Iris pumila*, habitat fragmentation, genetic polymorphism, Ukraine

Introduction

So far, only 3% of the territory of Ukraine remains fallow (Parnikoza et al. 2009). Of the 826 plant species listed in the Red List of Ukraine, 33.4% inhabit steppe biotopes. *Iris pumila* L., which is a characteristic species of the fescue-stipa (Cl. Festuco-Brometea Br.-Bl. et R.Tx.) steppes, is a non-Red List species that is still endangered. It is a typical steppe xerophyte of Ukrainian flora which habitat has been fragmented into multiple patches since the 18th century. The general population status of this species under the isolated conditions has not been studied in Ukraine. So the consequences of such isolation on its population genetics remain unknown.

Therefore, the goal of the present study was to address the consequences of isolation for *Iris pumila* L. populations from the perspectives of population ecology and genetics.

Material and Methods

General population assessment. A number of field trips took place during the 2010-2011 seasons to survey the populations that had been investigated before during 2000-2009. Published data also was used (Sykura and Shycha 2010, for review see Parnikoza et al. 2011). A number of populations were studied in the Dnipropetrovsk -2, Mykolaiv region – 3 and in the Crimea - 2.

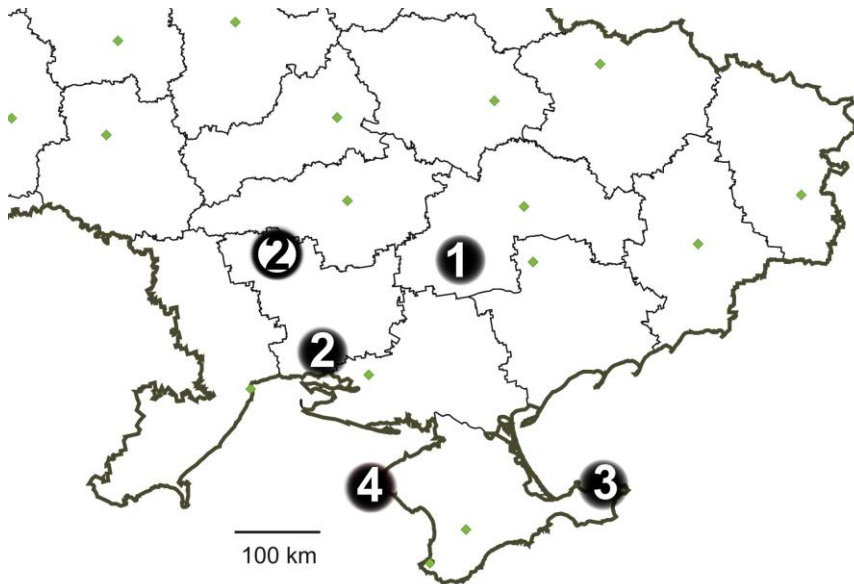


Figure 1. Geographic locations of the studied *Iris pumila* populations: 1 – Dnipropetrovsk region, 2 – Mykolaiv region, 3 – Kerch Peninsula (Crimea), 4 – Tarkhankut Peninsula (Crimea). Encircled is the location of the population near the Migia village studied by molecular genetics methods

Population size, fraction of ontogenetic stages, presence of successful flowering and fruiting were analyzed as described in Parnikoza et al. (2011). Special attention was paid to the effects of some adverse factors, such as burning, grazing, erosion, illegal collection of plants etc, on the populations.

Molecular genetics studies. For preliminary investigation eleven plants were used for molecular genetics analysis from a fragment of one population inhabiting petrophyle steppe areas near Migia which were the most numerous (over 1000 individuals) and well-studied on the population level. We mapped all analyzed individuals, including both mature generative and young pre-generative plants, on the area of 77 square

meters. We used five types of molecular markers dispersed over both coding and non-coding genome regions: nine RAPD (random amplified polymorphic DNA), eight ISSR (inter-simple sequence repeats), six RGAP (resistance gene analog polymorphism), four LP-PCR (long primer PCR – markers based on genes controlling abiotic stress response), and seven IRAP (inter-retrotransposon amplified polymorphism) primer pairs.

Results and discussion

The studied populations numbered hundreds (near Mykolaiv on the Alauda Peninsula and near the Kolarovo village, as well as on the Tarkhankut and Kerch peninsulas in the Crimea) to thousands (near Migia). Population densities ranged from 2 to 10 plants per square meter. The population numbers apparently depended on the size of the local steppe fragment. Thus, the studied population fragment near Migia (Mykolaiv region) was part of a large population inhabiting non-arable petrophyle steppe.

All the studied populations were dominated by mature generative plants, ranging from 95 to 100%. This data was on contrary with data from the Rostov region of Russia (Fedyeva et al. 2011) where populations of the species were balanced. In fact, mature generative plants may not flower in some years, but they differ from pre-generative individuals in that they have well-ramified systems of creepers and large beds. Flowering was observed in all of the populations, its presence, scale, and fruiting success being year-specific. For instance, any sign of successful fruiting was not found in some years (2005-2007) in the very dry plain steppes of the Kerch Peninsula in the Crimea. The dependence of fruiting on climatic conditions has been documented in multi-year studies of another meadow-steppe species – *Iris hungarica* (Waldst. et Kit) (Parnikoza et al. 2009). Given this dependence, the question of flowering and fruiting frequencies in *I. pumila* needs research on their population dynamics.

The studied populations are under profound human impact, the most extreme component of which being the habitat fragmentation. In particular, only solitary *I. pumila* plants were found in the Dnipropetrovsk region on tiny spots of virgin steppe along roadsides and river valleys. Undoubtedly, these are the most extreme forms of population existence.

Most populations of *I. pumila* suffer from the regular spring burning and grazing, which may restrict population renewal with juvenile individuals and, thus, promote the development of unbalanced populations dominated by mature generative plants. This scenario has been also demonstrated for other steppe perennials, like *Pulsatilla pratensis* Mill. and *Iris hungarica*, in

the Lysa Gora tract in Kyiv that is subject to regular burning (Parnikoza et al. 2007, 2009).

The sporadic character of seedling fixation may have also been caused by the unfavorable climatic conditions during the vegetative season, as well as the notable matting of the local soil surface.

The erosion of the sea and estuary coasts is an additional factor that affects the populations. In the vicinity of the Black Sea, the species inhabits narrow stretches along the eroded coasts that gradually collapse, which puts the existence of the species in peril. Other factors that negatively affect the existence of the species are steppe afforestation, junkyards scattered over steppe patches, mining operations, and illegal archaeology. Additionally, deliberate illegal collection of ornamentally valuable plants was noted. The effect of all these factors is particularly unpredictable given the absence of real population dynamics data for the species from most parts of its habitat. As the species is not included into the Red List of Ukraine, it is practically beyond the monitoring studies carried out by natural reserves staff and other research institutions of Ukraine. Under such circumstances, population shrinkage and loss of genetic diversity may remain unnoticed.

The indicative molecular genetic study of the fragment of *I. pumila* population near Migia revealed a high level of genetic polymorphism as inferred from the pooled data for all the markers. The polymorphism (P) was 78.4%, the average number of alleles per locus (A) – 1.78 ± 0.018 , the effective number of alleles per locus (A_e) – 1.33 ± 0.014 , Shannon index (S) – 0.33 ± 0.01 , Nei's gene diversity (expected heterozygosity H_e) – 0.208 ± 0.007 , and the Jacard genetic distances between plants (D_j) – 34.31–52.77% (mean genetic distance – 45.13%).

The high level of genetic polymorphism of the studied population fragment is in agreement with the species biology, e.g. cross-pollination and perennial life cycle, and suggests no immediate threat of its genetic decay. In the studied population fragment, sexual reproduction prevails over vegetative, as none of the samples were clones, even those growing in close proximity (the shortest distance between a pair of samples was 16 cm).

Thus, indicative genetic analysis suggests no signs of gene pool depletion in this isolated population of *I. pumila*. Notably, similar results have been obtained by Hungarian researchers for populations of the restricted-range steppe endemic *Dianthus diutinus* Kit. (Nemeth et al. 2011).

Therefore, the size and the balance of ontogenetic stages in populations of *I. pumila* primarily depend on the size of the steppe fragment. According

to the results within large steppe fragments, especially petrophyle patches, populations of this species can reach great numbers. Seed and vegetative renewal and bed outgrowth can be observed there. Similar results have also been reported for the Rostov-on-Don area in Russia (Fadyaeva et al. 2011). But the large area of the steppe remnant itself does not guarantee population success, as it can be opposed by severe human impact.

Still, we have only assessed a small population fragment near Migia. Comparison with other remote population fragments of various sizes and growing in different (e.g. dryer) conditions will demonstrate whether the genetic heterogeneity indices we obtained apply to other populations of the species.

Threats raised by the shrinking of the area of steppe fragments and the increased human impact make it necessary to consider inclusion of this species into the national Red List. This, among all, would promote inclusion of the species into national monitoring programs.

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