The effect of climatic factors on variation in leaf blade shape of *Globularia bisnagarica* L. (Plantaginaceae) within the East European fragment of the range.

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Introduction

The study of intraspecific morphological variability is one of the main goals of plant population biology. Determining the degree of intraspecific morphological variability at the population level, as well as identifying the factors responsible for its particular pattern can help in understanding the adaptive potential of a species. In present work, the authors made an attempt to reveal the main patterns of morphological variation in leaf blade shape of 25 populations of the rare, relict species *G. bisnagarica* L. (fig. 1) using geometric morphometrics approach.

Fig. 1. ***Globularia bisnagarica L.***

Sampling design

The sampling was carried out in 2021 from 25 *G. bisnagarica* populations located in the following subjects of Russian Federation: Saratov Region, Ulyanovsk Region, Samara Region, Orenburg Region, Republic of Bashkortostan, Republic of Tatarstan and Stavropol Territory (fig. 2).The studied populations were separated into six groups depending on the type and geographical location of the topographic features, to which their habitats are confined: P – Volga Upland, SYA – Sokskiye Yary, BB – Bugulma-Belebey Upland, OS – Obshchy Syrt, U – Southern Urals, and S – Stavropol Upland.

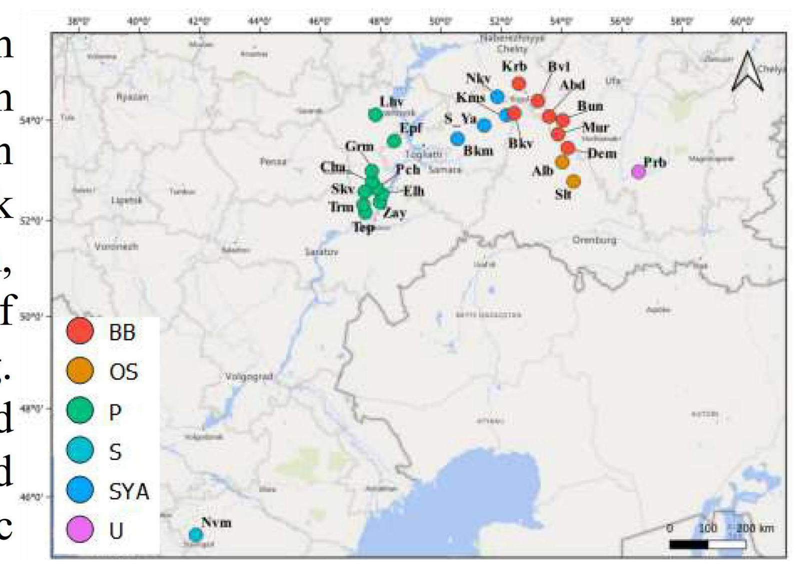

Fig. 2. Geographical location of studied *G. bisnagarica* populations.

Methods (digitizing)

From each population, one rosette leaf per individual were herbarized from 30 generative plants. Each specimen was photographed against a white background. Three fixed landmarks and two outlines were placed on the photographs, using the Tps series software. Landmark № 2 represented the leaf blade apex; № 1 and 3 were the points where the leaf blade starts to expand. Each outline consisted of 50 equidistant semilandmarks, limited by fixed landmarks at the ends (fig. 3).

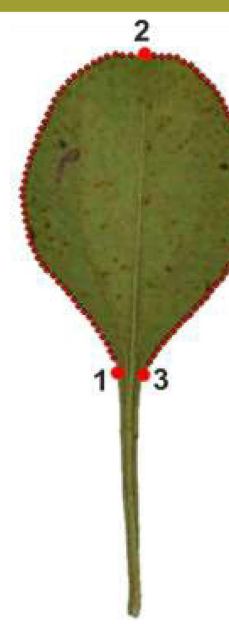

Fig. 3. Location of fixed landmarks and semilandmarks on a photograph of *G. bisnagarica* leaf blade.

Methods (statistical analysis)

Principal component analysis was used to identify the main patterns of morphological variation in leaf blade shape of *G. bisnagarica* populations. To investigate the relationships between the observed pattern and some environmental factors, which were represented by the most biologically meaningful climatic variables the multiple linear regression models was used. The mean populations scores of the first two principal components (PCs), reflecting the main patterns of morphological variation, were used as dependent variables in the modeling procedure. The predictors included the daily mean solar radiation in April, May, June and July, elevation, and 19 bioclimatic variables derived from temperature and precipitation, obtained from the WorldClim database with 30 s (1 km) spatial resolution. Due to the presence of multicollinearity in predictors, we used a selection procedure based on the values of the Variance inflation factor (VIF). The environmental variables with VIF values <3 were retained for subsequent analysis (tab. 1).

Table 1

Uncorrelated climatic variables from WorldClim database

Variable	Code
Maximum temperature of warmest month	Bio5
Mean temperature of wettest quarter	Bio8
Mean temperature of driest quarter	Bio9
Precipitation seasonality (coefficient of variation)	Bio15
Daily average solar radiation in July	Srad_07
Elevation	Elevation

The selection of optimal models was based on Akaike Information Criterion (AIC) values.

Results

The shape variation along the two first Principal Components: PC1 and PC2, explains 80.97% and 14.85% of the total variation, respectively, is associated with the length width ratio of the leaf blade, as well as with the narrowing place in its basal part (fig. 4). Thus, the shape changes from oblong, with a clear narrowing basal part, to a wider and spatulate with a smoother transitioning to a relatively wide basal part.

The optimal model for the geometric morphometrics PC1 included two predictors: precipitation seasonality and daily mean solar radiation in July. The optimal model for PC2 included: maximum temperature of warmest month, mean temperature of driest quarter, precipitation seasonality and daily mean solar radiation in July. The explained variance amounted to 52 % and 58 % respectively (tab. 2).

Results

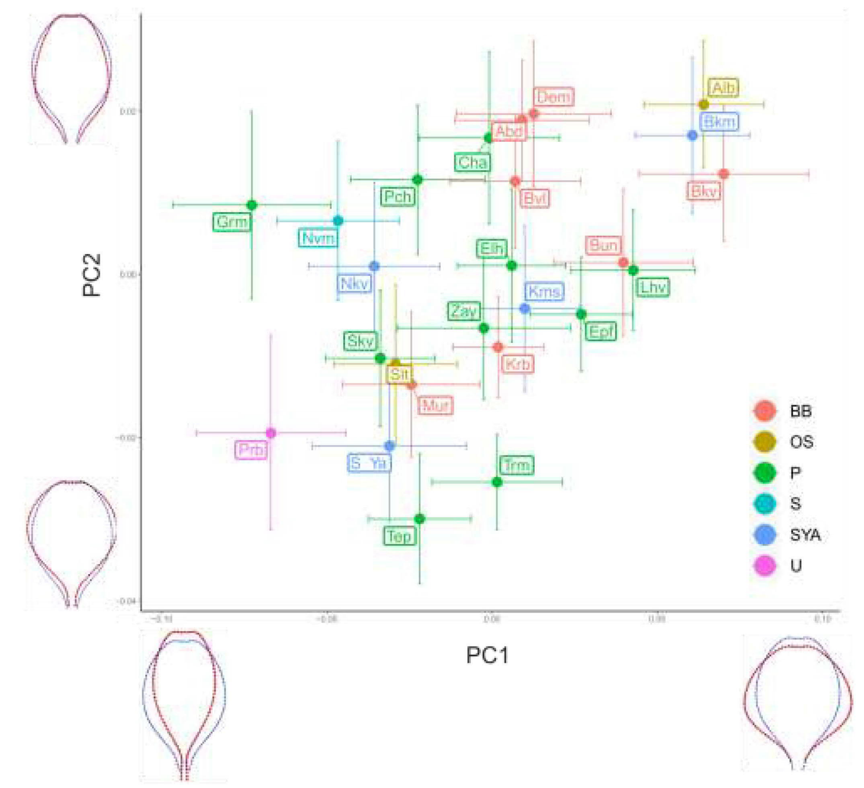

Fig. 4. Scatterplot obtained via Principal component analysis of *G. bisnagarica* leaf blade shape variables.

Table 2

Summary statistics for two full regression models containing all retained predictors (1, 3) and two optimal models with minimal AIC (2, 4)

Predictor	Dependent variable			
	PC1		PC2	
	1	2	3	4
Bio5	0.003		0.010**	0.011***
Bio8	0.007		-0.002	
Bio9	0.003		-0.009*	-0.007*
Bio15	-0.013	-0.014*	-0.005	-0.004
Elevation	-0.001		0.001	
Srad_07	-0.024**	-0.023***	-0.007*	-0.007**
R^2	0.538	0.522	0.589	0.578

Note: Significance codes: 0 '***'; 0.001 '**'; 0.01 '*'

Conclusions

Populations from the northeast and northwest of the study area were characterized by wide, spatulate leaf blades, while the plants from southern populations have the narrower and more elongated forms of leaf blades. The modeling results shown that substantial part of observed shape variation can be explained by the total influence of several climatic factors: precipitation seasonality, amount of solar radiation in July, maximum temperature of warmest month and mean temperature of driest quarter.

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