

Fitting the Morphological Diversity of *Poa* Sect. *Stenopoa* into a Taxonomic Framework

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Bluegrasses (*Poa* L.) of section *Stenopoa* is one of the most numerous, variable and intricate groups of grasses in temperate regions of Asia. The preliminary research of correlations and geographical distribution of 5 main discriminators have been undertaken. It has revealed 54 combinations of characters states (among 96 possible), but only 22 of them could be attributed to identified species. Significant correlation was found between ligule length and surface of rachilla. The divergence in geographical distribution of ligule length (along longitude) and lemma callus (along latitude) has been revealed. It confirms the morphological and geographical divergence and has indicated the main trend. The most morphological diversity has been revealed in Chinese provinces Sichuan and Yunnan.

Key words: biodiversity, *Poa* L., biogeography, Asia.

Bluegrass (*Poa* L., Poaceae) is one of the most common grasses, distributed in temperate and arctic-alpine regions¹, whereas the section *Stenopoa* represents one of the major and significant lineages. *Stenopoa* is distributed mainly Eurasia, but the center of its morphological diversity seems to be in Asia, the mountain territories seem to be the most rich². The species of this group are known to be the dominants of vegetative communities^{2,3}, therefore both the definition of their specific borders, and species number are of a great practice significance. Nevertheless, despite of having been researched during a long time, this group of grasses reminds to be one of the most difficult within *Poa*.

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The evolution of this section is considered to have gone on a way of xerophylisation and 4 levels or steps of xeromorphy (the position of uppermost node is a main marker of these levels) can be recognized among its species. Two main lineages, originated from mesomorphic *P. palustris* L. and *P. nemoralis* L. are suggested in this section^{2,4}. A small number of characters, suitable for discrimination, caused the additional difficulties. The study was focused on the qualitative characters. Unlike the quantitative ones, which variability quite often depends on environment, the qualitative features are more determined by genotype, therefore they can serve as markers for studying of evolutionary processes. Certainly, the fact, that same characters states may be caused by different evolutionary events, was taken into account, nevertheless, at studying of such large and difficult group as section *Stenopoa*, the revealing even the trends in

morphological and geographical divergence may be useful and productive. So, the phenogeographic researches of a large material are justified and allow make biogeographical assumptions. The main qualitative characters, used in *Stenopoa* taxonomy, are: position of uppermost node (indicate the level of xeromorphy and evolution, presented by 4 states), pubescence of lemma callus, rachilla and lemma between principal and secondary nerves (glabrous or pubescent), and ligule length of flag leaf (which frequently used in keys as quantitative character, presented by 4 states). Thus, on one side, five main characters, represented by 13 states, could be resulted in 96 possible combinations or morphs. On another side, the difficult geological history of Asia, numerous catastrophic changes of its terrestrial surface and its climate have resulted not only in unusual variety of landscapes and habitats, but also promoted high level of speciation and biodiversity. Indeed, the common and widespread species undergone the various changes and formed numerous morphs, deviated from types. Polyploidization, hybridization and apomixes, being common within *Stenopoa*, have caused the extraordinary polymorphism and made delineation of species and its taxonomy very difficult⁵. High polymorphism of the Bluegrasses is observed at the karyological level as well, and some of species are represented by several chromosome races^{6,7}. Thus, there is no clear notion on volume and the status of some members of this section.

At the same time it is necessary to notice, that the regular study of Asian flora has begun little more than 100 years ago, and, despite of Flora of USSR, Flora of Siberia, Flora of Russian Far East and most of volumes of Flora of China having been published, this region, being multifarious in the landscape and climatic relation, remains till now to be irregularly and insufficiently investigated in the botanical relation. Especially it concerns the out-of-the-way places of Central Asia. So, about 100 species were described within this section from Asian territory. Most of them are very similar morphologically, some of them were described without proper reason, based on the single herbarium sample, sometimes even without seeds, being, probably, germ-free. Because of high variability, many samples cannot be associated with any known species, dropping between 2 or even 3

ones. Indeed, from one hand, we can observe enormous biodiversity among species of *Stenopoa*, and from another hand, there are a lot of species, sometimes very similar and obscure.

In order to find out the taxonomical rank of numerous observed samples and their relationship, from the very beginning we should at least to find out at least the trends in distribution of the main character states (phenes), and correlations between them. Thus, the main goal of our preliminary research was 1) to reveal the correlation between main morphological characters, which are used for species discriminations in *Stenopoa*. 2) to find out the regularity in phenes distribution among I-III evolution groups. 3) to estimate the area within the studied territory, which is the most rich in *Stenopoa* morphs. Only I-III groups were involved in analysis, because of large size and extraordinary variability of the IV group. It will be researched separately. The proper classification and identification of this diversity, revealing of the status and relationship of observed samples is the goal of the next research.

MATERIALS AND METHODS

The research covered the South Siberia, Mongolia and China. This area is the most rich in different species of *Stenopoa* and its size is enough for biogeographical conclusions. Because of the majority of the materials, observed in Chinese herbaria, being supplied by labels only in Chinese, without of geographical coordinates, it was possible to identify only a province. The geographical coordinates of the central part of the provinces were used in the analyses. As far as the Chinese territory is large, such approach reflects the trend in geographical distribution of morphological patterns, and it seems to be justified for preliminary research.

Herbarium collections, obtained from BM, K, KW, L, LE, MHA, MO, MW, NS, NSK, PE, SSBG, TK, US, the personal collections, kindly represented by Dr. R. J. Soreng, as well as samples, collected by the authors during expeditions in Siberia and China, totally 1297 samples, have been used in the work. Four qualitative characters, listed above, were included in analysis. These characters were analyzed in total sampling and within 3 groups, selected in accordance with the level of xeromorphy.

The marker of this level is the fifth character – position of uppermost node (1– in upper 1/2; 2 – between 1/2 and 1/3; 3 – in lower 1/3 of stem length). For the visualization of distribution of the character states all observed combinations of mentioned above characters have been coded and then plotted on the map in accordance with their geographical position.

The correlations between characters were calculated by STATISTICA 7

Principle component analysis (PCA) and factorial analysis were performed by this program as well. The factorial analysis provides the possibility of studied objects space turning. Matrix turn allows arranging objects in such a manner that the main axis coincides with a direction of the most significant components that provides to evaluate the factors more easily⁸. In our case the using of quartimax row function has allowed to

increase the loadings on the first components and made the graph of characters dependence more evident.

RESULTS AND DISCUSSION

Five main characters, represented by 13 states, could form 96 possible combinations or morphs. The research has revealed 54 different combinations, but only 22 of them formally correspond to diagnoses of known species (Table 1). All other registered morphs (20.28 % of the investigated material), combined the characters of two or more different species. It confirms high polymorphism of section and may be caused by wide development of hybridization between species in section.

The stable species are known to have strong correlations between morphological

Table 1. Combinations of the basic morphological characters, appropriated to known species of sections *Stenopoa* (I-III evolutionary groups) in Asian Russia (Siberia), Mongolia and China

S. No.	Morph (species)	Position of upper most node (marker of evolution level)	Rachilla	Lemma callus	Lemma between veins	Ligule length	Number of observed samples
1	<i>P.nemoralis</i>	1	1	1	0	1	29
2	<i>P. palustris</i>	1	0	1	0	3	47
3	<i>P. urssulensis</i>	2	0	0	0	0	49
4	<i>P. urssulensis</i>	2	1	0	0	0	22
5	<i>P. urssulensis</i>	2	1	1	0	0	18
6	<i>P. urssulensis</i>	2	0	0	0	1	13
7	<i>P. urssulensis</i>	2	0	1	0	1	13
8	<i>P. urssulensis</i>	2	1	0	0	1	1
9	<i>P. urssulensis</i>	2	1	1	0	1	1
10	<i>P. faberi*</i>	2	0	0	0	4	113
11	<i>P.sphondylodes*</i>	2	0	1	0	4	271
12	<i>P. alta</i>	3	1	1	0	1	49
13	<i>P. nemoraliformis</i>	3	0	0	0	1	19
14	<i>P. hylobates</i>	3	0	0	0	4	46
15	<i>P. stepposa</i>	3	0	1	0	3	125
16	<i>P. ochotensis</i>	3	0	1	0	3	80
17	<i>P. reverdattoi</i>	3	1	0	1	1	0
18	<i>P. relaxa</i>	3	0	1	0	1	19
19	<i>P. varia</i>	3	0	1	0	4	80
20	<i>P. orinosa</i>	3	1	0	0	3	6
21	<i>P. orinosa</i>	3	1	0	0	4	8
22	<i>P. orinosa</i>	3	1	1	0	3	19

Position of uppermost node: 1- in upper 1/2; 2 – between 1/2 and 1/3; 3 – in lower 1/3. Rachilla: 1- glabrous; 2- pubescent. Lemma callus: 1- glabrous; 2- with the tuft of long hairs. Lemma between veins: 1- glabrous; 2- pubescent. Flag leaf ligule length: 1- less 1 mm; 2 - 1-2mm; 3-2-3 mm; 4 more than 3 mm. *Poa faberi* differs from *P. sphondylodes* in much softer leaves and habitat.

characters, gained during evolution, whereas the hybridization results in the destroying of these correlations. R.L. Berg⁹ and I.I. Shmalgauzen¹⁰ have shown, that intraspecific divergence from the very beginning capture the correlations between the characters, and only then concern the characters. Indeed, being in integrated complex, every character can vary only in the limits of correlations, which was stated as adaptive result of functional system. Hence, the divergence of correlations between characters among the same species may be caused by genetic differentiation and can detect the beginning of the evolution process. Therefore the analysis of correlation between characters represents the certain interest for study of microevolutionary processes.

The mapping of the main character's combinations has demonstrated some trends in their distribution. The samples with glabrous callus seem to be concentrated mainly in the western part of the China, especially in province Xizang (Tibet), and it was observed mainly within Bluegrasses of II and III evolutionary levels. In order to confirm or contradict this view, the research of the correlations between separate morphological characters, and also latitude and longitude, has been undertaken. It has revealed regularity in the frequency of some characters on different evolutionary level and in different parts of investigated area (Tables 2, 3).

The most correlation was observed between ligule length and rachilla pubescence. As it was mentioned above, these characters are the

Table 2. Correlations between separate states of morphological characters, latitude and a longitude (above diagonal – the correlations of integrative sampling, under diagonal – correlations within I level)

	1	2	3	4	5	6	7
1	1	-0.08	-0.08	0.11	0.09	0.06	-0.06
2	-	1	0.12	-0.39	0.04	0.05	0.06
3	-	0.10	1	0.03	-0.05	0.15	0.39
4	-	-0.44	-0.05	1	0.02	-0.35	-0.01
5	-	*	*	*	1	-0.01	-0.02
6	-	0.13	0.04	-0.38	*	1	0.18
7	-	-0.07	0.37	0.14	*	0.11	1

* The samples with lemma pubescent between veins were not registered among I level.

1 – Position of uppermost node. 2 – Rachilla pubescence. 3 – Surface of lemma callus. 4 – Flag leaf ligule length. 5. – Lemma between veins. 6 – Latitude. 7 – Longitude.

Table 3. Correlations between separate states of morphological characters, latitude and a longitude (above diagonal - the correlations in II level-group; under diagonal - correlations in III level-group)

	1	2	3	4	5	6
1	1	0.02	-0.24	-0.01	-0.05	0.02
2	0.15	1	0.24	-0.05	0.09	0.45
3	-0.39	-0.07	1	-0.01	-0.38	0.16
4	0.06	-0.06	0.06	1	0.06	0.03
5	0.02	0.24	-0.25	-0.06	1	0.12
6	0.13	0.33	-0.23	-0.04	0.29	1

* The samples with lemma pubescent between veins were not registered among I level.

1 – Position of uppermost node. 2 – Rachilla pubescence. 3 – Surface of lemma callus. 4 – Flag leaf ligule length. 5. – Lemma between veins. 6 – Latitude. 7 – Longitude.

main discriminators between basic species *P.nemoralis* and *P.palustris*, which than gave rise two lineages (Tables 2, 3). The correlation between these characters reach its greatest values (-0.44) among mesomorphic samples (I level), then it decrease till -0.24 among xeromesomorphic samples (II level), and then rises again till -0.39 among xeromorphic samples (III level). In integrative sampling this correlation is also high (-0.39). Such a high correlation between these characters in I group may be caused by stability of morphological structure of *P.palustris* and *P.nemoralis*. It seems to be reduced within *Purssulensis* Trin., which is supposed to be of hybrid origin, then the correlations are partially restored at *P. nemoraliformis* Roshev., *P. hylobates* Bor and *P. stepposa* (Kryl.) Roshev., which are known to be quite stabile species. At the same time, quite low correlation between these characters testify against genetical integrity of system and may be an evidence of hybridization and high evolutionary potential of studied group. Other morphological characters are not correlated significantly.

The correlations of morphological characters with longitude or latitude indicate the geographical divergence and shift. Both latitude and altitude demonstrated high correlations with some morphological characters. The latitude is significantly negatively correlated with ligule length. The coefficients are quite high in integrative sampling and in different evolutionary groups, the lowest is in III. (Tabl. 2, 3). It may be caused by distribution of species with short ligule (*P.nemoralis*, *P. nemoraliformis* and partially *Purssulensis*) mainly at the North of area, whereas the species with very long (3,5-5 mm) ligule –

P.hylobates and *P.sphondylodes* Trin – occur at the South. In the III group latitude is connected quite closely with surface of lemma callus as well. The longitude significantly correlated with surface of lemma callus. The most correlation (0.45) was observed in the II group, whereas the weakest one was in III group (Tabl. 2, 3). Such a situation may be explaining by absents of the morphs with glabrous callus among the mesomorphic samples (I level).

The Principal component analysis was undertaken to reveal the possible irregularity in the distribution of samples on the plot, caused by morphological or geographical reasons. PCA has not revealed regularity in the cases distribution (Fig. 1A). Some discrete clouds along the II PC are caused by grouping samples while indicated geographic coordinates for the most of Chinese samples. As it was mentioned above, in the most of cases it was possible to identify the province only, and it caused such a discrete distribution of the most Chinese material. The PCA has not revealed special features in geographical distribution of the different evolution (xeromorphic) groups. All of them are scattered on the plot homogenously (Fig. 1 A). The factorial analysis, using function quartimax row, has shown, that it was the longitude, which demonstrated the great influence (together with surface of lemma callus) on the II PC (Table 4, Fig. 1B). The first PC is caused mainly by ligule length and rachilla pubescence. It confirms, that the influence of the main characters, which differ the basic mesomorphic species *P.palustris* and *P.nemoralis*, keep their significance as discriminators at the II and III level

Table 4. Loading on the major factors, obtained from of factorial analysis, using quartimax row function

Characters	Factor I	Factor II	Factor III	Factor IV
Position of uppermost node	-0,34	-0,18	-0,63	0,37
Rachilla pubescence	0,77	0,11	0,18	0,22
Surface of lemma callus	0,02	0,83	0,02	-0,01
Flag leaf ligule length	-0,84	0,087	0,27	0,19
Surface of lemma between veins	0,04	-0,00	0,01	0,92
Latitude	0,33	0,22	-0,77	-0,15
Longitude	-0,01	0,81	-0,12	0,00
Expl.Var	1,51	1,46	1,11	1,06
Prp.Totl	0,22	0,21	0,16	0,15

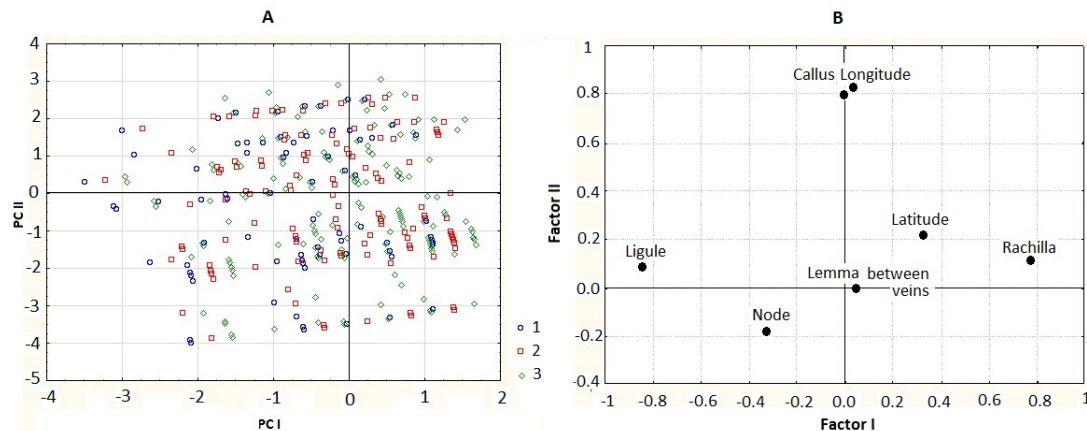


Fig. 2. Multivariate analysis of the main characters of *Stenopoa* distribution.
A – Principal components analysis. B – Factorial analysis (function quartimax raw).

Thus, some shifts in geographical distribution of ligule length, rachilla pubescence and surface of lemma callus have been inferred both from research of numerous collections and from analyses. The morphological differentiation is known to be the consequence of two reasons: adaptations to an environment (and it is well interpreted when the character is of strongly pronounced adaptive value) or it may be caused by genetic reasons only, for example, resulted from hybridization in the part of its area with any species, which poses new character state. In that case it can be treated as evolutionary event, as far as it is caused by changing at the genetic level. Since all of these 3 characters have no obvious adaptive value, we can assume the evolution process in this group, caused mainly by hybridization.

The glabrous lemma callus in Western part of area might be obtained due to hybridization with growing there *P.asperifolia*. Abnormally narrow spikelets, glumes and lemma, and the superlong ligula, exceeding 3.5-5 mm, which are common among *P.asperifolia*, *P.hylobates* and *P.faberi*, may be the additional evidence of hybridization as well. The long ligule, exceeding 3.5 mm, does not meet in Siberia at all, being occasionally shown only at separate individuals of hybridogenous *P. ochotensis* in the Far East. Further, at the South-East, it occurs at xeromesomorphic *P.sphondylodes*, which range reaches Taiwan. At the western part of area such a long ligule was registered among xeromesomorphic samples of the West of Tibet, Sichuan and Yunnan,

and among mesomorphic *P.faberi* and its hybridogenous forms. The appearance of such a character state in the different parts of area may be caused by different events, but other methods should be used for checking of these hypotheses. The xeromorphic samples with pubescent rachilla are more common in southwest part of China, occurring in Eastern Siberia and NE of China. It should be noticed, that in China it shows weak correlations with other characters, whereas in Siberia and Mongolia pubescent rachilla usually associated with short ligule and other morphological characters, typical for *P.nemoralis* at the I level and *P.alta* Hitch. at the III one. The study has shown, that *P.alta*, being quite stable in Siberia, in China turns to vary very much even within the same population (or herbar sheet). In the West, in Chinese province Xinjiang, pubescent rachilla is quite common, combining with short ligule. It may be caused by influence of *P.nemoralis* or its derivatives.

CONCLUSION

The research has revealed, that due to excessive variability, a lot of samples of Asian *Stenopoa* could not be assigned to any known species. Fifty four combinations of 13 states of 5 morphological characters have been observed among the taxon under consideration, but only 22 morphs could be at least formally attributed to identified species. All other registered morphs combine the characters of different species and

may be of hybrid origination. Significant correlations were revealed only between ligule length and surface of rachilla, it was kept on all three levels, but the most pronounced in among mesomorphic species. Decrease of its value among II group may be explained by hybrid origination of *Purssulensis*, inferred from many other features. Both latitude and longitude correlate closely and on the all levels with ligule length and surface of lemma callus respectively. Such a correlation indicate the shift and divergence in geographical distribution of mentioned phenes. The most morphological diversity has been revealed in Chinese provinces Sichuan and Yunnan, which are rich in different ecological niches.

Besides of a lot of samples being with mixed characters, the most of investigated populations proved to be mixed as well, and contained the forms, which combined the character states of two or three species. The study using the methods of traditional morphology has indicated that in *Poa*, especially in its section *Stenopoa* we deal not so much with species, but rather with complexes of different genesis and different structure, which consist of several ecological geographic races and hybrid populations of unknown taxonomical rank, rather than with species, traditionally accepted by botanists. The finding out the rank of observed morphes, expose their relationship and origination is a target of the next research. The proper taxonomic interpretation of this diversity will be based on data, obtained from molecular methods.

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