

ORIGINAL ARTICLE

Reproduction Strategies of Clonal Plants of *Potentilla conferta* in Uzbekistan and Mongol

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Abstract

Clonal plants combine sexual and clonal reproduction, which contribute differently to plant fitness. Reproductive analyses have highlighted the importance of clonal growth in shaping the spatial genetic structure in *Potentilla conferta* Bunge, a herbaceous rhizomatic clonal distributed in hot sand dunes. We investigated the reproduction system of *P. conferta* at two populations in Mongol and three natural populations in Uzbekistan. The measurements of 19 quantitative or qualitative morphological characters were taken on each of total individuals directly from their natural habitats. Some morphological characteristics between Mongolia and Uzbekistan populations showed a slight heterogeneity of variance. Especially, the length of internodes (LFI and LSI) and characteristics of root (LLR and NOR) were shown a significant difference between two countries ($P < 0.05$). *P. conferta* of Uzbekistan has most ramets at short distance intervals 30~100 cm. In light conditions, *P. conferta* of Uzbekistan was significantly less resilience than *P. conferta* of Mogol. In drought conditions, although there was not shown significant difference, *P. conferta* in Uzbekistan was less resilience than that in Mogol. The core analysis indicates that *P. conferta* in Uzbekistan is the more resistant than that of Mongol and seems to do by sexual reproduction strategy during several strong environmental disadvantages such as drought events.

Key Words : Clonal characteristics, *Potentilla conferta*, Mongol, Uzbekistan

1. Introduction

Some of the world's most extensive deserts are in Asia, from the dune of the Arabian Peninsula to the high and cold desert steppes of Mongolia. Even when sparsely vegetated, some of these harsh environments support surprising botanical diversity. Desert plants will often bloom only once or twice a year for a short time (Ali et al., 2000). Their seeds remain dormant until it rains. Rainforest plants often bloom continuously and produce seeds that start growing immediately.

Morphological differentiation of species within their geographic distribution has been of considerable interest in the study of the evolution of species. Morphological data from variables measured on a continuous scale are important in generating and testing evolutionary hypotheses and taxonomic hierarchies. Divergence among populations may occur as a result of microevolutionary changes in isolated populations in different nature in time (Silander and Antonovics, 1979). These changes in isolated populations in different environments produce individuals with different ecological

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tolerances to environmental factors, resulting in the differentiation of divergence ecotypes. The actual geographical distribution of a species may be reflective of these changes over geological time. Differentiation between populations has received much attention in recent years using different approaches and many distinct taxa (Hamrick et al., 1992).

Flowers are very useful for identifying plants and providing aesthetic pleasure for humans, but they have a more vital function—they are the sexual reproductive organs of plants. Many plants also have methods of asexual reproduction (vegetative reproduction), which produces offspring that are genetically identical to the parent: root-sprouting (limberbush, palo verde, aspen), stolons and rhizomes (agaves, strawberries, many grasses), and aerial plantlets (some agaves, mother-of-millions, kalanchoe). All of the progeny of asexual reproduction are clones of their parent plants. (A clone is a group of organisms that are genetically identical; in the case of flowering plants each clone originates from a single seed.). Clonal structure in clonal plants can affect sexual reproduction. Individual ramets can decrease reproduction if their neighbors are ramets of the same genet due to inbreeding depression or self-incompatibility. Clonal structure in plants can dramatically influence the mating system, favoring either outcrossing and selfing. The interaction of sexual reproduction and clonality affects both population genetic diversity and individual plant fitness (Handel, 1985; Tarasjev et al., 2012).

Many plants, especially rhizomatous and stoloniferous species, have physical connections among ramets although the level of persistency is highly variable among species and habitats (Sobey and Barkhouse, 1977). Studies on the genetic structure of clonal plant populations have received increased interest over the past decade with the advent of electrophoretic

techniques, which allow us to better access the genotypic composition of populations (Clegg, 1980; Cook, 1983). Clearly, descriptive genetic work on both sexual and asexual plant populations is needed as well. Despite the importance of genetic variation data and population genetic structure of these clonal species, detailed studies of the levels and distribution of genetic variation have not been performed on most species in Mongol and Uzbekistan, and are particularly lacking for plants with both sexually- and asexually-reproductive ability (Huh, 2001).

Located in the landlocked plateau of Central Asia between China and Russia, Mongolia covers an entire area of 1,566,500 km² it takes the 15th place with its size in the world. Across the eastern part of the country stretches the vast land grasslands of the Asian steppe. The annual precipitation amount is low, averaging 200-220 mm and ranging from less than 50 mm per year in the extreme south (Gobi and desert region) to 400 mm per year in limited areas in the north.

Uzbekistan is located in central Asia, bounded on the north and west by Kazakhstan, on the east by Kyrgyzstan and Tajikistan, on the south by Afghanistan, and on the southwest by Turkmenistan. Uzbekistan has an area of 447,400 km², which is slightly smaller than California. Uzbekistan's area includes 22,000 km² of inland water, mainly the Aral Sea. It is one of only two countries in the world bounded only by other landlocked countries. Although the Fergana Valley receives just 100 to 300 mm of rainfall per year, only small patches of desert remain in the center and along ridges on the periphery of the valley. The Uzbekistan geography is dominated by its vast stretches of desert land. About eighty percent of the geography of Uzbekistan is dominated by desert

areas. The Qizilkum Desert is one of the main desert, which lie in between Samarkand and Bukhara.

Potentilla conferta Bunge was distributed over the extremely continental climate with harsh environmental conditions. These species are long-lived herbs, which can reproduce extensively by vegetative rhizomes and potentially by sexually produced seeds. Rhizomes are generally prostate stems at the nodes.

In this study we investigated the reproduction system of three natural populations of *P. conferta* in Mongol and two populations in Uzbekistan using clonal characteristics. Consequently, there is a need to study the investigation of the nature to better understand the morphological characteristic of genus *Potentilla* species. The objective of this paper is to; 1) compare the mode of reproduction for two different desserts, 2) and analyse morphological variation within and among populations.

2. Materials and Methods

2.1. Collecting sites

P. conferta was collected from three populations which represent the geographical range and have effective large population-sizes in Uzbekistan on July 2012. Because Plants of this species were not distributed within communities but, rather, were clustered in distinct patches (Table 1). *P. conferta* was also collected from two populations in Mongol. Table 1 lists the collection sites used in this study, along with three-letter abbreviations, and population sizes.

Quadrat sampling is based on measurement of replicated sample units referred to as quadrats or plots. Eleven contiguous 1 m x 1 m quadrates were established in a transect 11 m long. The distance between selected individuals was about 10 m in order to avoid including individuals with common lineage (Fig. 1).

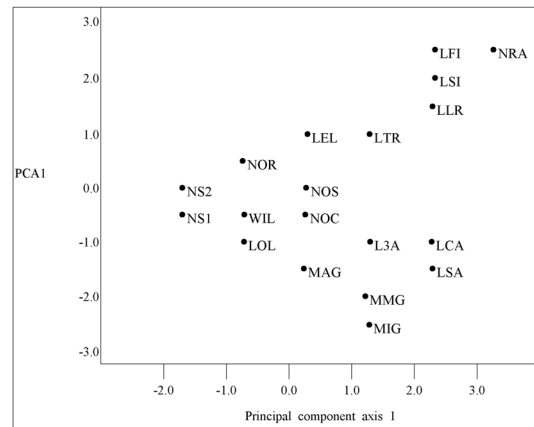


Fig. 1. Plots of factor loadings for variables obtained by principal component analysis (PCA) of the both *Potentilla* species. Abbreviations are shown in Table 2.

2.2. Morphological characteristics

The measurements of quantitative characters and observation of qualitative characters were taken on each of total individuals directly from their natural habitats. Table 2 was shown lists of the 19 morphological characteristics. The measurements made on plants were arranged in a data matrix indicating the means of plant characteristics for each population.

Table 1. Numeric code, population location, and sample sizes of *Potentilla conferta*

Code	Location	No. of subpopulations	Sample size (n)
PBM-1	Zavkhan aimag, Mongol	5	35
PBM-2	Terelj, Mongol	4	30
UZB-1	Bukhara, Uzbekistan	3	27
UZB-2	Samarkant, Uzbekistan	4	30
UZB-3	Aidargkul, Uzbekistan	4	40

Table 2. List of 19 descriptive characteristics used in the morphological analysis

Acronym	Characteristic derivation	Unit or Category
LOL	Number of leaf	ea
WIL	Width of leaf (maximum)	mm
LEL	Length of leaf	mm
LTR	Ratio of length/width (leaf)	ratio
LCA	Length from basal sinus to central lobe apex	mm
LSA	Length from basal sinus to second lobe apex	mm
L3A	Length from basal sinus to 3rd lobe apex	mm
MAG	Major groove of leaf	mm
MMG	Middle groove of leaf	mm
MIG	Minor groove of leaf	mm
NS1	Number of serrulate at palmatifid (1st margin)	ea
NS2	Number of serrulate at palmatifid (2nd margin)	ea
NRA	Number of ramets	ea
LFI	Length of the first internode	cm
LSI	Length of the second internode	cm
LLR	Length of the longest root	cm
NOR	Number of roots	ea
NOS	Number of stamen	ea
NOC	Number of capel	ea

2.3. Data analysis

The main trends in variation between localities (populations) were summarized using univariate and multivariate analyses. Measurements of each particular trait were compared using one-way analysis of variance (ANOVA) with site of origin as a factor. Whenever a significant difference was found ($P < 0.05$), multiple comparison tests were performed following SYSTAT procedures (Zar, 1984). Bartlett's test was applied to test the homogeneity of variance and transformations of variables were performed when data were not homoscedastic.

Species diversity may be thought of as being composed of two components (Ludwig and Reynolds, 1988). The first is the number of species in the community, which ecologists often refer to as species richness. The Shannon-Weiner index of diversity was used to represent species richness. The second component is species evenness or equitability.

Pearson correlation coefficients were calculated using the Statistical Analysis System to determine correlations between morphological characteristics.

We investigated the relationships between size, reproduction, and clonal growth in *P. conferta* under a range of conditions, using the simple model (Loehle, 1987; Weiner, 1988) for the size of sexual structures

$$r = b_r \cdot (v - a_r)$$

and the size of clonal structures

$$c = b_c \cdot (v - a_c)$$

where v is the vegetative size of the primary ramet, a_r and a_c are the minimum sizes required for sexual reproduction and clonal growth, respectively, and b_r and b_c are the slope parameters for the two relationships.

To study resilience and resistance, we used variation in precipitation in the form of drought events and light as external perturbations (DeClerck et al., 2006).

The data were standardized for each characteristic, with the raw data matrix transformed such that each characteristic had a mean of zero as a standard deviation. Principal component analyses (PCA) were

carried out to determine how the various isolates and populations of the *P. conferta* relate to each other. In addition, we conducted to detect differences among populations considering several characters simultaneously of variances using the Statistical Analysis System (Tilman and Lehman, 2001).

Pair-group analysis was utilized using an unweighted method (UPGMA) of agglomerating clustering based on Euclidean distance similarity matrix (Felsenstein, 1993). This cluster analysis was performed with morphological phenogram among populations based on quantitative variation.

3. Results

The major morphological characteristics of *P. conferta* held in general for polypetalous with yellow flower, 5 sepals, 5 petals, many stamens and carpels. The analysis of variance, calculated from 19 morphological characteristics in each population, showed a slight heterogeneity of variance (Table 2). 19 morphological characteristics of *Potentilla* species between Mongol and Uzbekistan showed a slight heterogeneity of variance. For example, eleven characteristic values were homoscedastic, of which ten values departed significantly from zero ($P < 0.05$). The length of internodes (LFI and LSI) were shown a significant difference between two species ($P < 0.05$). The length from basal sinus to central lobe apex (LCA) has the highly significant positive correlations with the length of from basal sinus to second lobe apex (LSA), and 3rd lobe apex (L3A).

PCA was conducted to detect differences between Mongol and Uzbekistan based on the 19 morphological characteristics. In the two-dimensional plot constructed based on the morphological data derived from the PCA (Fig. 1), all the 19 characteristics are clearly separated into five or six classes. LCA, LSA, L3A, AP1, AP2, AP3, and ACL are classified one group. MAG, MIG, and MMG are also classified one

group. LOL, WIL, NS1, and NS2 are consisted of one group. LEL and LTR are same trend. LFI and LSI are formed a long ellipse. NRA showed a clear separation of the other characteristics. Principal components 1, 2, and 3 contained 62.0%, 45.6%, and 13.2% of the total variance, respectively. The major first components were the length of internodes, the number of ramets, and width between two lateral lobe apex. The second components was mostly concerned with length of leaf (LEL and LTR).

The number of ramets in Mongolia *P. conferta* decreased with increasing geographic distance from viviparity (Fig. 2). However, Uzbekistan *P. conferta* has most ramets at short distance intervals 30~100 cm.

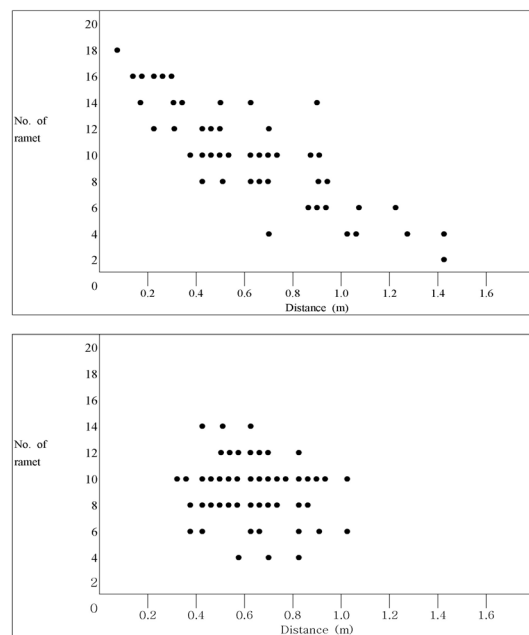


Fig. 2. The number of ramets according to spatial distribution, Upper: Mongol. Lower: Uzbekistan.

Shannon-Wiener functions differed significantly among populations ($F = 6.487$, $p < 0.05$), with Mongolia *P. conferta* having significantly higher value (2.105) than that of Uzbekistan (1.874) (Table 3). The richness indices were also shown a significant differences

between two deserts. However, the evenness indices were not a significant differences between two deserts.

Table 3. Diversity indices of Shannon-Weiner, richness, and evenness

Parameters	Location		
	Mongol	Uzbekistan	
Diversity	Mean	2.105	2.077
	Range	2.005 - 2.205	2.054 - 2.114
Richness	Mean	1.994	1.208
	Range	1.780 - 2.207	1.032 - 1.361
Evenness	Mean	0.961	0.916
	Range	0.958 - 0.964	0.896 - 0.935

The relationship between clonal growth and size was only slightly weaker than that between sexual reproduction and size (r^2 in Table 4). Because of the minimum size of for sexual reproduction but not clonal growth, the ratio reproductive size to clonal structure size increased with plant sizes.

Though response trends in resilience to drought and light events followed the different patterns regard of drought severity and light conditions. In light conditions, *P. conferta* in Uzbekistan was significantly less resilience than that of Mongol. In drought conditions, although there was not shown significant

difference, *P. conferta* in Mongol was less resilience than that of Uzbekistan.

The cluster analysis is concordant with the results obtained from the morphological data. The isolates of *P. conferta* from Mongol and Uzbekistan were separated in two clusters: Uzbekistan and Mongol. Clustering of *P. conferta* populations, using UPGMA, was performed based on morphological characteristics (Fig. 3). Many of the morphological characteristics studied among the populations were associated with latitude. For example, the more northern Mongolia population group, the less the lobe of leaf but more serrulate at palmatifid. However, most morphological variation except LOL and LTR is significantly correlated with latitude.

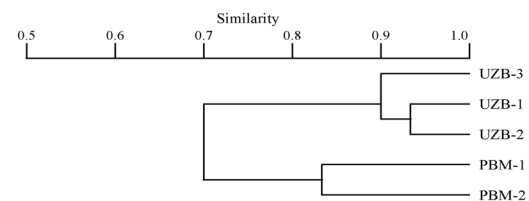


Fig. 3. A dendrogram showing the phenetic relationships among the three populations of Uzbekistan and two populations of Mongol based on data of morphological traits. Codes of populations are given in Table 1.

Table 4. Parameter estimates for the relationship between the size of sexual or clonal structures and the sizes of vegetative structures at harvest for seed- and rhizome-derived plants of *Potentilla conferta*

Region and mode	Minimum size (x-intercept±SE)	Slope (±SE)	r^2
Sexual structures			
Mongol			
Seed-derived plants	4.67±1.28	0.48±0.05	0.80
Rhizome-derived plants	6.56±1.36	0.36±0.03	0.74
Uzbekistan			
Seed-derived plants	5.68±2.31	0.32±0.02	0.66
Rhizome-derived plants	4.54±1.23	0.27±0.02	0.49
Clonal structures			
Mongol			
Seed-derived plants	-5.44±3.69	0.45±0.03	0.69
Rhizome-derived plants	-2.38±1.56	0.12±0.01	0.67
Uzbekistan			
Seed-derived plants	-12.52±5.44	0.13±0.02	0.61
Rhizome-derived plants	-3.82±3.73	0.20±0.03	0.75

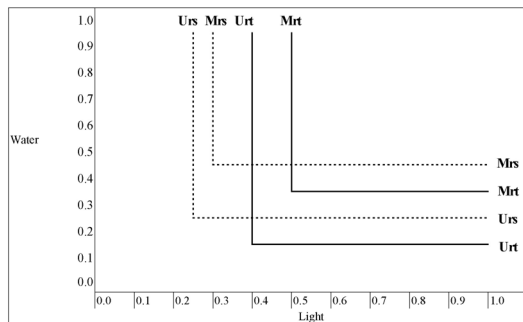


Fig. 4. Zero net growth isoclines based on each species' limitation for light and water. Urs (···) and Urt (—) are the resilience and resistance of *P. conferta* for light and water in Uzbekistan, respectively. Mrs (···) and Mrt (—) are the resilience and resistance of *P. conferta* for light and water in Mongol, respectively.

4. Discussion

Sexual reproduction is generally the main source of genetic variation in populations and clonality can potentially reduce outcrossing rates and the formation of new genetic combinations (Charpentier, 2000; Honnay and Bossuyt, 2005).

The larger number of populations per species did not differ from each other in most morphological characteristics. However, some morphological characteristics of two deserts were distinct. Although some morphological variations were within populations, one of the most striking features of this study was existed a significant difference between deserts. The length of internodes, and number of ramets were those which were considered likely to be useful in the subsequent intensive examination of *P. conferta* (Figs. 1 and 2).

Phenotypic plasticity is the ability of an organism to change its phenotype in response to changes in the environment (Price et al., 2003). Because of the limited number of plants available, phenotypic plasticity could not be assessed quantitatively in the present study. Phenotypic of quantitative traits are subject to environmental influence, *P. conferta*

showed the plastic response to environmental variations. The heterogeneous nature of fragmentation among populations ought to be an obvious indicator of the need to protect the remaining desert vestiges.

P. conferta is ecologically restricted, growing in warm, dry, and high pressure atmospheric conditions in Mongol. Therefore, local populations are isolated each other, and they are discretely distributed. Furthermore, as discussed above, each local population is subdivided, consisting of many subpopulations. Species with a relatively narrow niche, and with discrete, isolated populations ("habitat specialists") like *P. conferta*.

In most species of *Potentilla*, especially perennial herbs, species consist of a series of internodes (Eriksen, 1997; Spommer, 1999). Each ramet may produce only one terminal flower in the year it is formed (Cook, 1983; Hartnett and Bazzaz, 1985). Many plants have two reproductive strategies, sexual reproduction via seeds and clonal propagation via the development of vegetative rhizomes through the growth of a coordinated group of cells that form a meristem (Holsinger, 1991). *P. conferta* is no exception, and consists of repetitive units (ramets) which may be interconnected via rhizomes. The species flowers mainly in April or May, producing many inflorescences per ramet (cyme), although infrequently one inflorescence per ramet is produced. We also observed that fruits (achene) start to be visible in July. These observations suggest that the present populations might have been founded from asexual fragmentation and dispersal of preexisting clones rather than from sexually-produced seed.

Interestingly, plants have evolved unique alternative life cycles that bypass typical seed production in favor of clonal reproduction systems. This may seem counter intuitive because sexual reproduction should lead to greater genetic diversity in offspring compared to clonal plants (Hamrick et al., 1992). However, investing in clonal reproduction seems to increase the

likelihood that a species can colonize specific environmental niches. It has been observed that many of the perennial species in a given ecosystem tend to combine both sexual and clonal forms of reproduction (Ellstrand and Roose, 1987). *P. conferta* usually propagates by asexually-produced stolens when several strong environmental disadvantages influence the habitat of this species. The species has physical connections among ramets (Fig. 2). Their asexual reproduction assures the stabilization and persistence of a phenotype that is well adapted to the immediate environment (Templeton, 2002). Although *P. conferta* is able to reproduce by sexually-produced seeds, its ratio of asexual/sexual reproduction has not yet been studied. These results suggest that *P. conferta* in Uzbekistan has larger number of ramets than that of Mongol. Thus asexually-production by stolon is dominant in Uzbekistan *P. conferta* populations. In addition, it cannot rule out the possibility that sexual reproduction occurs at a low rate. Mongolia forests grow under the Central Asian condition with an extreme climate. Across the eastern part of the country stretches the vast land grasslands of the Asian steppe. Although the annual precipitation amount is low. However, Mongol occurs much precipitation in Winter with snow than Uzbekistan. We can suppose that Uzbekistan has more extreme environmental condition for *P. conferta* than Mongol. It might contribute the growth of *P. conferta* in Mongol and the long rhizomatous and nodes. *P. conferta* in Uzbekistan has short rhizomatous and nodes with many flowers for the survival (Fig. 2 and Table 4). This analysis indicates that *P. conferta* in Uzbekistan is the more resistant species than that of Mongol and usually propagates by clonal growth during several strong environmental disadvantages such as drought events (Freestone, 2006). Due to the virtually unsolvable Aral Sea problem, high salinity and contamination of the soil with heavy elements are especially widespread in Karakalpakstan, the

region of Uzbekistan adjacent to the Aral Sea. The bulk of the nation's water resources is used for farming, which accounts for nearly 84% of the water usage and contributes to high soil salinity. Water deficit and salt stresses are global issues to ensure survival of agricultural crops and sustainable food production (Jaleel et al., 2008).

5. Conclusions

The 19 morphological characteristics of *Potentilla* species between Mongol and Uzbekistan showed a slight heterogeneity of variance. In the two-dimensional plot of PCA, all the 19 characteristics are clearly separated into five or six classes. The number of ramets in Mongolia *P. conferta* decreased with increasing geographic distance from viviparity. However, Uzbekistan *P. conferta* has most ramets at short distance intervals 30~100 cm. Many of the morphological characteristics studied among the populations were associated with latitude. For example, the more northern Mongolia population group, the less the lobe of leaf but more serrulate at palmatifid. Though response trends in resilience to drought and light events followed the different patterns regard of drought severity and light conditions.

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