



Full Length Article

Fertility Variation in Wild Rose (*Rosa canina*) over Habitat Classes

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ABSTRACT

Number of fruits (female fertility), stamens (male fertility) and filled seeds were assessed in wild rose (*Rosa canina* L.) populations sampled from Beysehir watershed divided into two main (A & B) and six sub-habitats classes based on ecological characters. Fertility variation, effective number and coancestry were estimated based on fecundity assessment in the populations. Beside, the habitats were compared by multiple analyses of variance. Results showed that main habitats (A & B) had similar mean and coefficients of variation for the assessments. The mean values among sub-habitats within the main habitat 'A' varied considerably unlike the main habitat 'B' which was more homogenous. Coefficient of variation of fecundity estimates among individuals was more than 60% in main habitats and most of the sub-habitats. Variation was the lowest for seed number in main and sub-habitats and the highest for female flower production in main and sub-habitats (with the exception of one sub-habitat). Coancestry computed with total gene pool of flowers was higher than that for number of filled seeds in both main and sub habitats. Statistically significant ($p \leq 0.05$) differences were found among habitat classes for flower and filled seed productions. Results emphasized that habitat class was more effective on production of studied characters than that of fertility variation. © 2011 Friends Science Publishers

Key Words: Dog rose; Effective number; Fruit; Sibling coefficient; Site class; Stamen

INTRODUCTION

Wild rose, also known as 'dog rose' (*Rosa canina* L.), is a hermaphroditic shrub that occurs throughout Turkey, individually or in groups. The species is self-compatible but their large flowers are designed for pollination by insects and out-crossing is probably common (Uggla, 2004). The fruits and flowers are used as food, in cosmetics and pharmaceutical industries (User, 1967). The species is also used in landscape planning and erosion control (Ilisulu, 1992). It keeps its hips during the winter, so this species has an important role for wildlife, and biological diversity. Rose has also valuable essential oil used in a wide range of many industries (Khan & Rehman, 2005).

Fecundity and fertility variation have an important role in selection and management of gene conservation areas, and maintenance of genetic diversity. Estimation of fertility is one of the important tools for tree breeding, gene conservation and seed production programs (Kang *et al.*, 2003; Bilir *et al.*, 2005) and in evolution and genetic management of populations (Bila, 2000). Cones, flowers, pollen, fruits and seeds have been used to estimate fertility in plants (Roeder *et al.*, 1989; Xie & Knowles, 1992; Savolainen *et al.*, 1993). The counting of female and male reproductive structure is a quick and inexpensive method to estimate fertility differences among parents (Kang &

Lindgren, 1999; Bila, 2000). Many studies have been carried out recently on fertility variation in forest tree populations (Bila & Lindgren, 1998; Bila *et al.*, 1999; Bilir *et al.*, 2003 & 2005; Kang *et al.*, 2003; Varghese *et al.*, 2006; Varghese *et al.*, 2008; Nicodemus *et al.*, 2009). Fertility data of shrubs and hermaphroditic species are however very limited.

Genetic structure of individuals/populations (e.g., phenotype, fertility) and habitat characters (e.g., altitude, breeding zone & seed transfer region) are used in selection and management of gene conservation areas. The main objective of this study was to investigate fecundity and fertility variation with habitat class and determine their role in gene conservation and management of the species.

MATERIALS AND METHODS

Site description and habitat classification: The sampled watershed is located between latitudes 37°26' to 38°03' N and longitude 31°15' to 31°46' E and has a mean altitude of 1120 m. It includes the second largest lake (Beysehir Lake) of Turkey. The altitude reaches up to 3000 m. at Dedegul Mountain. The Mediterranean climate and Interior Anatolian climate prevails in the watershed. According to Thornthwaite classification, the low lying regions of the watershed have semi-drought, semi-humid and humid

climate, mesothermal, close to oceanic effect with the strong water deficit in summer. The most widespread trees are Crimina juniper (*Juniperus exelsa*), Crimean pine (*Pinus nigra*), Cupper oak (*Quercus cerris*), Lebanon Cedar (*Cedrus libani*), Cilician Fir (*Abies cilicica*) (Ozkan, 2003).

In this study, the watershed was divided into two main (Sultan Mountain - 'A' & Dedegul Mountain - 'B') and six sub-habitats based on geographical and ecological characters proposed by Ozkan (2003). The characters included landform, climate (annual temperature, minimum & maximum temperature, annual rainfall, humidity, seasonal wind direction, climate type), main rock and vascular plant species (distribution of tree, shrub & herb species).

Main habitat 'A' has mostly limestone based soils, while main habitat 'B' is composed mostly by alluvial deposits, marly and marly limestone and schistes. The main habitat 'B' is affected by north and east air masses coming from the Beysehir Lake. Hence, main habitat 'B' has more humid climate than 'A'. Also, main habitat 'A' is composed of A-I, A-II and A-III sub-habitats, semi drought and cool conditions in A-I; drought and cool/semi warm conditions in A-II and semi drought and semi warm conditions in A-III characterized, while main habitat 'B' is composed of B-I, B-II and B-III sub-habitats, characterized with semi-humid and semi-warm, humid/semi humid and semi-warm, humid/semi humid and cool conditions, respectively (Ozkan, 2003).

Flowering data: Sixty individuals were sampled from each sub-habitat of natural occupation of the species without any replication in the watershed. Number of fruits (female fertility) was counted in the sampled individuals in each population in July and August, 2006. Three fruits were collected from each radial directions and the number of stamens (male fertility) was counted in each fruit. Total number of stamens per individual was estimated using the average number of stamens per fruit and the total number of fruits per individual.

Seed data: Seeds were extracted from the fruits and immersed in water for 12-16 h for separating the empty seeds. Total number of filled seeds per individual was estimated using the average number of filled seeds per fruit and the total number of fruits per individual. Pollination and seed production of the species occur in the same year.

Fertility variation: Fertility was defined as the relative proportion of fertile individuals (i.e., contribution) to the entire population. The female and male fertility of an individual is considered to be proportional to the number of female and male gametes produced by the individual (Gregorius, 1989). The female fertility variation (ψ_f) and male fertility variation (ψ_m) were estimated as (Kang & Lindgren, 1999):

$$\psi_f = N \sum_{i=1}^N f_i^2; \psi_m = N \sum_{i=1}^N m_i^2 \quad [1]$$

Where N is the census number, f_i is the female fertility and m_i is the male fertility of the individual i .

Sibling coefficient, (Ψ , called total fertility variation) is the probability that two genes randomly drawn from the same parent, is used describe fertility variation as compared to equal contribution from all individuals. Sibling coefficient was estimated as described by Kang and Lindgren (1999):

$$\Psi = N \sum_{i=1}^N \left(\frac{f_i + m_i}{2} \right)^2 = N \sum_{i=1}^N p_i^2 \quad [2]$$

Where N is the census number, f_i is the female fertility, and m_i is the male fertility of the individual i , p_i is the probability that two genes in the offspring come from the same parent i .

Fertility variation was also estimated based on the number of filled seed (ψ_s) as:

$$\psi_s = N \sum_{i=1}^N s_i^2 \quad [3]$$

Where N is the census number, s_i is the fertility for filled seed production of the individual i .

Effective parent number and coancestry: The effective numbers of female [$N_{p(f)}$] and male [$N_{p(m)}$] parents, total gametic gene pool [N_p] and filled seed [$N_{p(s)}$] were estimated based on census number (N) and fertility variation of female (ψ_f), male (ψ_m) and gametic gene pool (Ψ) and seed number (ψ_s) by Kang and Lindgren (1999) as:

$$N_{p(f)} = N/\psi_f; N_{p(m)} = N/\psi_m; N_p = N/\Psi; N_{p(s)} = N/\psi_s \quad [4]$$

Group coancestry (Θ) is the probability that two genes chosen at random from a gene pool are identical by descent (Cockerham, 1967). Group coancestry (Θ_ψ) was estimated considering male and female fertility (Lindgren & Mullin, 1997) as:

$$\Theta_\psi = 0.5 \sum_{i=1}^N \left(\frac{f_i + m_i}{2} \right)^2 = 0.5 \sum_{i=1}^N p_i^2 \quad [5]$$

Where N is the census number, f_i is the female fertility and m_i is the male fertility of the individual i , p_i is the probability that two genes in the offspring come from the same parent i .

Group coancestry was estimated considering fertility variation in seed production (Θ_{ψ_s}) as:

$$\Theta_{\psi_s} = 0.5 \sum_{i=1}^N s_i^2 \quad [6]$$

Where N is the census number, s_i is the fertility, based on filled seed production of the individual i .

Comparison of sub-habitats: Sub-habitats were compared by multiple analysis of variance (MANOVA) in following

linear model and grouped by Duncan's multiple range test (Duncan, 1974) in SPSS package program.

$$Y_{ijk} = \mu + S_i + M_j + e_{ijk}$$

Where Y_{ijk} is the observation from k^{th} individual of the j^{th} sub-habitats of the i^{th} main-habitat, μ is overall mean, S_i is the random effect of the i^{th} sub-habitat, T_j is the effect of the i^{th} main habitat and e_{ijk} is random error.

RESULTS

Flower and seed production: There were large differences in the number of fruits, stamens and filled seeds among sub-habitats within and between main habitats, while they were similar in the main habitats. The number of female flowers ranged from 110 (B-II) to 195 (B-III) and male flowers from 2197 (B-II) to 4178 (B-III). The number of seeds ranged from 1598 (A-I) to 3346 (B-III) (Table I). Statistically significant ($p \leq 0.05$) differences were found between main habitats and among sub-habitats for flower and filled seed productions by results of analysis of variance. The sub-habitats had two homogeneous groups for the characters (Table II).

There was significant positive correlation ($p=0.001$, $r>0.98$) between female and male flower production in all main and sub-habitats. Coefficient of variation was very high both within and among habitats for reproductive traits. Coefficient of variation was higher for female fecundity compared to male fecundity and filled seeds per tree in all main and sub-habitats. The sub-habitats of main habitat 'A' were more homogenous than that of main habitat 'B' (Table I).

Fertility variation, effective number of parents and coancestry: Fertility variation, effective number of parents and coancestry were very low for the reproductive traits and habitats (Table III). There were small differences between main habitats in fertility variation. The trend was the similar for effective parent number and coancestry (Table III). But, the difference was larger among sub-habitats than main habitats especially in main habitat 'A'. Fertility variation based on seed number was low in all habitats and generally higher for female fecundity. Effective parent number was very low ($\approx 50\%$) for most of the characters and habitats (Table III).

DISCUSSION

The differences in average number of fruits, stamens and filled seeds were larger among sub-habitats than main habitats (Table I). It was also well in accordance with results of analysis of variance and Duncan's multiple range test (Table II). The difference showed the effect of sub-habitats on reproductive characters. Several environmental factors such as temperature, light and nutrition are known to play an important role in floral initiation and development, but the

mechanism involved is not well understood (Kinnaird, 1992). Differences in age and environmental variation, mainly in soil properties, are known to influence fruiting and seed set in different populations in natural forests (Bila & Lindgren, 1998). In my study, while climate (e.g., humidity) and soil characters (e.g., main rock) differed between main habitats (Ozkan, 2003), there was only a small difference in fecundity (Table III). The main habitat B has a more humid climate than 'A'. The environment can affect the female and male fertility differently. In monoecious species pollen production is reported to dominate on dry sites, while seed and fruit production are expressed more on wetter sites (Freeman *et al.*, 1981). This report tallies well with the results of the current study where habitat 'B', which has a more humid climate (Ozkan, 2003), produced more flowers and seeds than main habitat 'A' (Table III).

In wind pollinated species temperature, wind speed, rain during pollen dispersal, and the physical distribution of inter-pollinating individuals are considered important factors for successful pollination (Sedgley & Griffin, 1989). In insect pollinated species, rain, temperature and wind influence the foraging behavior of many pollinators (Sedgley & Griffin, 1989). 'A' significant phenotypic correlation ($p=0.001$, $r>0.98$) was found between number of fruits and number of stamens in all habitats (Table I). 'A' similar trend was reported in an earlier study on the number of female and male flowering in different plant species such as by Bilir *et al.* (2005), Varghese *et al.* (2008) and Nicodemus *et al.* (2009). Coefficient of variation was very high within and among habitats for the reproductive traits (Table I). It showed effect of genetic structure and habitat characters on reproductive characters. Coefficient of variation for the assessments were the highest in female and it was ordered from high to low as female, male and seed number in all main and sub-habitats (Table I). Cones, flowers, pollen, fruits and seeds have been used to estimate fertility in plants (Roeder *et al.*, 1989; Xie & Knowles, 1992; Savolainen *et al.*, 1993). But, filled seed is the last stage of fertilization, so number of filled seeds gives an accurate estimate of female fertility variation. The female fertility variation (ψ_f) and male fertility variation (ψ_m) were also related to coefficients of variation in female (CV_f) and male (CV_m) flower production among individuals by Kang and Lindgren (1999) as:

$$\psi_f = CV_f^2 + 1; \psi_m = CV_m^2 + 1$$

Fertility variation, effective number and coancestry were similar in main and sub-habitats implying that they were independent of ecological differences or habitats. Bilir *et al.* (2005) reported that there was no relation between altitude and fertility variation in natural populations of *Pinus brutia*.

Female fertility variation was higher than that of male in the main habitats and most of the sub-habitats (Table III). An opposite trend was reported in natural forest tree stands

Table I: Averages, standart error, and coefficient of variation in the number of female and male flowers and the number and weight of filled seed in the habitats class

Main and sub habitats	Average± standart deviation				CV (%)		
	female	male	filled seeds	r*	female	male	filled seeds
A-I	126±16.3	3010±464.5	1598±301.1	0.985	95.4	83.6	68.5
A-II	161±20.8	3134±405.6	2717± 667.8	0.996	61.6	57.9	52.5
A-III	186±24.3	3844±492.6	2637± 376.7	0.987	98.7	100.7	90.3
Mean-A	157±15.0	3329±324.3	2317± 276.1	0.983	78.2	76.6	62.6
B-I	171±12.9	3738±282.1	2498±272.5	0.997	109.7	104.3	95.9
B-II	110±20.1	2197±462.6	1676±335.9	0.990	109.2	100.5	79.4
B-III	195±25.2	4178±537.6	3346± 536.5	0.989	100.1	100.3	80.5
Mean-B	158±11.8	3371±261.3	2506±234.5	0.991	100.0	96.3	79.8

*; r is the phenotypic correlation coefficient between female and male fertility production, statistically significant at least 0.01 probability level for all values in the table

Table II: Results of Duncan’s multiple range test

Female flowering		Male flowering		Number of filled seeds	
Population	Homogen. group*	Population	Homogen. group	Population	Homogen. group
B1	a	B1	a	A1	a
A1	ab	A1	ab	B1	a
A2	ab	A2	ab	B2	ab
B2	ab	B2	b	A3	ab
A3	c	A3	b	A2	ab
B3	c	B3	b	B3	b

*; the same letter/s were not significantly different at $p \leq 0.05$

Table III: Fertility variation for female and male flower production (ψ_f & ψ_m), total gene pool (Ψ), seed number (ψ_s) and effective parent number for female and male flower production ($N_{p(f)}$ & $N_{p(m)}$), gametic gene pool (N_p), seed number ($N_{p(s)}$), and coancestry for gene pool (Θ_Ψ) and seed number (Θ_{ψ_s}) in the habitats

Habitats	Fertility variation				Effective parent number*				Coancestry	
	ψ_f	ψ_m	Ψ	ψ_s	$N_{p(f)}$	$N_{p(m)}$	N_p	$N_{p(s)}$	Θ_Ψ	Θ_{ψ_s}
A-I	1.91	1.70	1.84	1.47	31.4 (0.52)	35.3 (0.59)	32.7 (0.54)	40.8 (0.68)	0.015	0.012
A-II	1.38	1.34	1.36	1.28	43.5 (0.73)	44.9 (0.75)	44.0 (0.73)	47.0 (0.78)	0.011	0.011
A-III	1.97	2.01	1.99	1.82	30.4 (0.51)	29.8 (0.50)	30.1 (0.50)	33.0 (0.55)	0.017	0.015
Mean-A	1.61	1.59	1.61	1.39	111.7 (0.62)	113.4 (0.63)	111.9 (0.62)	129.3 (0.72)	0.005	0.004
B-I	2.20	2.01	2.16	1.92	27.4 (0.46)	28.7 (0.48)	27.7 (0.46)	31.2 (0.52)	0.018	0.016
B-II	2.19	2.01	2.13	1.63	30.0 (0.50)	29.8 (0.50)	28.1 (0.47)	36.8 (0.61)	0.018	0.014
B-III	2.00	2.01	2.01	1.65	27.2 (0.45)	29.9 (0.50)	29.9 (0.50)	36.4 (0.61)	0.017	0.014
Mean-B	2.00	1.93	1.98	1.64	90.4 (0.50)	93.4 (0.52)	91.0 (0.51)	110.0 (0.61)	0.006	0.005

*; relative effective parent number ($=N_p / N$) in parentheses

(Kang *et al.*, 2003). Large differences in fertility among trees were reported in natural populations of forest tree species (Shea, 1987; Bila & Lindgren, 1998; Kang *et al.*, 2003; Nicodemus *et al.*, 2009). It was concluded based on a survey that Ψ of a magnitude up to 3 could be typical in natural populations (Kang *et al.*, 2003). In my study it was lower than 3 in all main and sub-habitats. Gene diversity (GD) in seed crop was estimated based on effective number (N_p) or coancestry (Θ) as: $GD=1-0.5/N_p=1-\Theta$ by Kang and Lindgren (1998). Gene diversity is 1 for an ideal population. High genetic diversity is important for natural regeneration/sustainability (e.g., adaptation to global warming) of a species. The loss in GD in the studied habitats is estimated to be less than 5% (Table III). The accumulation of relatedness (Θ) was the lowest in A-II (0.011) and the highest in B-I (0.018).

In conclusion, habitat class could be used as a criterion to obtain economically high fruit crop based on differences among habitat classes. Low effective number of parents in some habitat classes could be balanced by different treatments such as tending. Similar fertility variations in female, male and filled seeds showed that one of the characters could be used for fertility estimation in the species. Collection female/fruit data was easier and cheaper than that of others. So, number of fruit could be used in estimation of fertility variation.

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REFERENCES

- Bila, A.D., 2000. Fertility variation and its effects on gene diversity in forest tree populations. *Ph.D Thesis*. Swedish University of Agricultural Science, Umeå, Sweden
- Bila, A.D. and D. Lindgren, 1998. Fertility variation in *Milletia stuhlmanii*, *Brachystegia spiciliformis*, *Brachystegia bohemii* and *Leucaena leucocephala* and its effects on relatedness in seeds. *For. Genet.*, 5: 119–120
- Bila, A.D., D. Lindgren and T.J. Mullin, 1999. Fertility variation and its effect on diversity over generations in a Teak plantation (*Tectona grandis* L.f.). *Silvae Genet.*, 48: 109–114
- Bilir, N., K.S. Kang and D. Lindgren, 2003. Fertility variation and effective number in the seed production areas of *Pinus radiata* and *Pinus pinaster*. *Silvae Genet.*, 52: 75–77
- Bilir, N., K.S. Kang and D. Lindgren, 2005. Fertility variation in six populations of Brutian pine (*Pinus brutia* Ten.) over altitudinal ranges. *Euphytica*, 141: 163–168
- Cockerham, C.C., 1967. Group inbreeding and coancestry. *Genetics*, 56: 89–104
- Duncan, A.J., 1974. *Quality Control and Industrial Statistics*. Richard D. Irwin Inc. Homewood, Illinois
- Freeman, D.C., E.D. MacArthur, K.T. Harper and A.C. Blauer, 1981. Influence of environment on floral ratio of monocious plants. *Evolution*, 35: 194–197
- Gregorius, H., 1989. *Characterization and Analysis of Mating Systems*. Ekopan Verlag, Germany
- Ilisulu, K., 1992. *Medical and Spices Plants*. Ankara University, Agricultural Faculty, Ankara, Turkey
- Kang, K.S. and D. Lindgren, 1998. Fertility variation and its effect on the relatedness of seeds in *Pinus densiflora*, *Pinus thunbergii* and *Pinus koraiensis* clonal seed orchards. *Silvae Genet.*, 47: 196–201
- Kang, K.S. and D. Lindgren, 1999. Fertility variation among clones of Korean pine (*Pinus koraiensis* S. et Z.) and its implications on seed orchard management. *For. Genet.*, 6: 191–200
- Kang, K.S., A.D. Bila, A.M. Harju and D. Lindgren, 2003. Fertility variation in forest tree populations. *Forestry*, 76: 329–344
- Khan, M.A. and S.U. Rehman, 2005. Extraction and analysis of essential oil of *Rosa* species. *Int. J. Agric. Biol.*, 7: 973–974
- Kinnaird, M.F., 1992. Phenology of flowering and fruiting of an east African riverine forest ecosystems. *Biotropica*, 24: 187–194
- Lindgren, D. and T.J. Mullin, 1997. Balancing gain and relatedness in selection. *Silvae Genet.*, 46: 124–129
- Ozkan, K., 2003. Properties and classification of sites on Beyşehir. *Ph.D Thesis*. Istanbul University, Forestry Faculty, Istanbul, Turkey
- Nicodemus, A., M. Varghese, B. Nagarajan and D. Lindgren, 2009. Annual Fertility Variation in Clonal Seed Orchards of Teak (*Tectona grandis* L.f.) and its Impact on Seed Crop. *Silvae Genet.*, 58: 85–94
- Roeder, K., B. Devlin and B.G. Lindsay, 1989. Application of maximum likelihood methods to population genetic data for the estimation of individual fertilities. *Biometrics*, 45: 363–379
- Savolainen, O., K. Karkkainen, A. Harju, T. Nikkanen and M. Rusanen, 1993. Fertility variation in *Pinus sylvestris*: a test of sexual allocation theory. *American J. Bot.*, 80: 1016–1020
- Sedgley, M. and A.R. Griffin, 1989. *Sexual Reproduction of Tree Crop*. Academic Press Ltd, London
- Shea, K.L., 1987. Effects of population structure and cone production on outcrossing rates in Engelmann spruce and subalpine fir. *Evolution*, 41: 124–136
- Uggla, M., 2004. Domestication of wild roses for fruit production. *Ph.D. Thesis*. Swedish University of Agricultural Science, Sweden
- User, E.T., 1967. An investigation for C vitamin and traditional uses on wild rose occupy in Turkey. *Turkish J. Hygiene*, 27: 39–60
- Varghese, M., A. Nicodemus, B. Nagarajan and D. Lindgren, 2006. Impact of fertility variation on gene diversity and drift in two clonal seed orchards of teak (*Tectona grandis* Linn. f.). *New For.*, 31: 497–512
- Varghese, M., R. Kamalakannan, A. Nicodemus and D. Lindgren, 2008. Fertility variation and its impact on seed crops in seed production areas and a natural stand of teak in southern India Impact of fertility variation on gene diversity and drift in two clonal seed orchards of teak (*Tectona grandis* Linn. f.). *Euphytica*, 160: 177–187
- Xie, C.Y. and P. Knowles, 1992. Male fertility variation in an open-pollinated plantation of Norway spruce (*Picea abies*). *Canadian J. For. Res.*, 22: 1463–1468

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