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DETERMINATION OF POLLEN VIABILITY AND *IN VITRO* POLLEN GERMINATION OF *ROSA DUMALIS* AND *ROSA VILLOSA*

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Abstract

Pollen quantity, viability and germination of four genotypes of *Rosa dumalis* and *R. villosa* were investigated. The number of anthers per flower were 90.3 in genotype 4 (*Rosa villosa*) and 116.4 in genotype 1 (*Rosa dumalis*). Genotype 1 showed the highest pollen viability on the basis of both TTC and IKI tests.

Most of the rose species are diploid but there are several species with higher ploidy levels as well, especially tetraploids and also pentaploids, in section *Caninae*. Diploid species are usually self-sterile whereas the polyploids are self-compatible. The deviating section of *Caninae* contains mostly pentaploid species and is characterized by the *Caninae* meiosis with an uneven chromosomal contribution from the two parents. In addition, these species are self-fertile, sometimes apomictic, and have reduced pollen viability but a high seed set (Nybom *et al*. 2005).

In general, there is a linear relation between pollen viability and germination capability in many fruit species (Grigs *et al*. 1971). Germination capability of pollen depends on various factors, namely nutrition conditions of species and varieties used and environmental factors (Eti and Stosser 1988). To investigate pollination potential, estimates should be made of pollen quantity and viability, as well as of pollen germination capability.

This study was designed to determine the qualitative and quantitative characteristics of the pollen and to assess *in vitro* pollen germination of some rose genotypes that are presently being domesticated in Turkey.

In the present study, four genotypes from *Rosa* section *Caninae* were used belonging to *Rosa dumalis* (genotypes 1 and 2) and *Rosa villosa* (genotypes 3 and 4). The number of anthers per flower were determined by counting anthers of 40 flowers from each genotype and the number of pollen per anther was determined by hemacytometer with six replications (Eti 1990). Pollen viability was tested according to Eti (1991) in 1% TTC (2,3,5-triphenyl tetrazolium chloride) and IKI (iodine+potassium iodide). The morphological homogeneity level of pollen was also investigated according to Eti and Stosser (1988). For *in vitro* pollen germination, 5, 10, 15, 20, 25, 30, 35 and 40% sucrose and 0.03, 0.01 and 0.1% boric acid were used in hanging drop method. In addition 1% agar + 15% sucrose combinations in Petri dishes were also used. The experiment was set according to the randomized block design with four replications (each replication includes 50 pollen) and the values were evaluated by the DMRT. Data were arc-sine transformed for analysis but non-transformed means are presented.

There were statistical differences ($p < 0.01$) among genotypes in terms of the number of anther per flower, the number of pollen per flower, the number of pollen per anther, pollen homogeneity and pollen viability. Average anther number in a flower and pollen numbers per anther was highest in genotype 1 as 116.4 and 1617, respectively (Table 1). The highest pollen number per flower was 188.219 in genotype 1 and followed by the genotype 2 (*R. dumalis*) as 154.157. The genotype 3 and 4 (*R. villosa*) had the lowest pollen numbers with 127.075 and 102.400, respectively (Table 1). Gunes *et al*. (2005) reported that the number of anther per flower of rose species were between 81.4 (*R. villosa*) and 148.1 (*R. elliptica*) similar to present result.

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However, the number of pollen per flower and the number of pollen per anther in different *Rosa* species (Gunes *et al*. 2005) were lower than the present study. A lot of factors such as species, cultivars, age of plants, nutritional conditions, culture environment affect the amount of pollen production in fruit species (Eti 1991).

Table 1. Number of anthers and pollen, and pollen viability in rose genotypes.

Genotype	Anther per flower	Pollen per flower	Pollen per anther	Pollen viability (%)	
				TTC	IKI
1	116.4 a **	188.219 a **	1617 a **	47.24 a**	48.36 a**
2	114.7 a	154.157 b	1344 ab	43.15 b	42.41 b
3	92.0 b	127.052 c	1381 ab	33.90 c	34.20 c
4	90.3 b	102.400 d	1134 b	31.88 c	33.30 c

**Means with different alphabet in a vertical column are significantly different at 0.01 level

Both the *R. dumalis* genotypes had higher pollen viability than *R. villosa* genotypes. This could be the effect of species. Genotype 1 (*R. dumalis*) had highest pollen viability in both the tests which are 48.36% in IKI and 47.24% in TTC. The results of both TTC and IKI tests indicated poor pollen viability of both the species namely 31.8-47.2% in TTC and 33.30-48.36% in IKI. It was previously reported that pollen viability of *R. canina, R. dumalis, R. rubiginosa* and *R. villosa*, all belonging to section *Caninae*, varies considerably among species and pollen viability was recorded between 23 and 45% (Werlemark 2000, Ueda and Akimoto 2001).

The pollen morphological homogeneity was higher in *R. dumalis* genotypes (57.02-59.33%) than *R. villosa* genotypes (51.35-53.87%), respectively (Fig. 1). Jicinska *et al.* (1976), reported that morphological homogeneity of *R. canina* pollens varied between 29.6 and 71.2%.

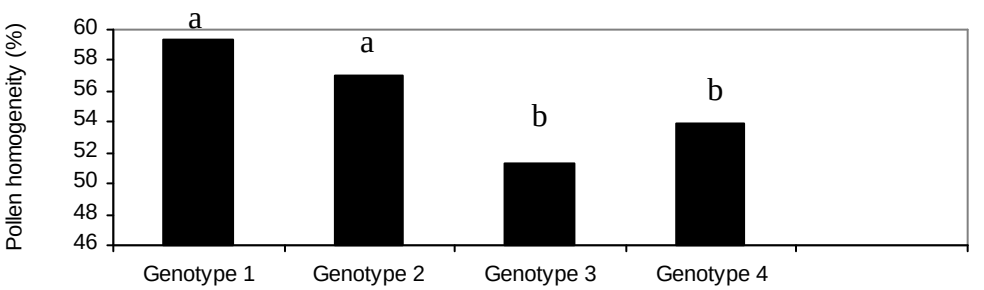

Fig. 1. Pollen homogeneity of rose genotypes.
**Means with different alphabet in a vertical column are significantly different at 0.01 level.

In the hanging drop method, none of the pollens of all the genotypes germinated in distilled water (control treatments). Pollen germination was increased with tbe increase of sucrose concentrations up to 35% in germination medium for *R. dumalis* and 30% for *R. villosa* genotypes (Table 2). The highest pollen germination percentage (32.04-36.25) was observed in *R. dumalis* genotypes at 35% sucrose medium and 17.42-19.01% in *R. villosa* genotypes at 30% sucrose medium (Table 2). Koncalova (1975) reported higher pollen germination percentage of *R. hugonsis* at 30 and 35% sucrose concentrations which supports the present findings.

Among boric acid treatments, 0.01% was found to be more effective for *R. dumalis* and 0.03% for *R. villosa* genotypes. The highest pollen germination was obtained from genotype 1 (*R. dumalis*) with 32.12% and followed by genotype 2 (*R. dumalis*) with 29.22% (Table 2).

Table 2. The effect of different media on percentage pollen germination in Rosa genotypes.

Medium	Genotype 1	Genotype 2	Genotype 3	Genotype 4
Distillate water (control)	0.00 d**	0.00 c**	0.00 b**	0.00 b**
Sucrose 5%	0.00 d	0.00 c	0.00 b	0.00 b
Sucrose 10%	10.00 cd	9.00 bc	4.71 ab	6.35 ab
Sucrose 15%	11.71 cd	9.09 bc	7.12 ab	8.27 ab
Sucrose 20%	15.13 c	15.68 bc	8.37 ab	11.13 ab
Sucrose 25%	17.89 bc	16.76 b	18.91 a	16.34 a
Sucrose 30%	26.25 b	28.34 a	19.06 a	17.42 a
Sucrose 35%	36.25 a	32.04 a	6.67 ab	13.12 a
Sucrose 40%	28.32 ab	30.27 a	1.41 ab	4.73 ab
Boric acid 0.01%	15.40 bc	16.88 ab	9.74 ab	6.36 ab
Boric acid 0.03%	14.12 cd	11.18 bc	8.99 ab	10.02 ab
Boric acid 0.1%	10.00 cd	8.97 bc	0.00 b	3.04 ab
Agar 1%+Sucrose 15%	32.16 ab	29.22 a	8.00 ab	11.21 ab

**Means with different alphabet in a vertical column are significantly different at 0.01 level.

Based on the present research findings it may be concluded that pollen production, viability and germination capacity varied within and between *Rosa* species. One of the reasons different ploidy levels of the species used.

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