International journal of Agronomy and Plant Production. Vol., 4 (2), 314-318, 2013
Available online at http:// www.ijappjournal.com
ISSN 2051-1914 ©2013 VictorQuest Publications

Comparison of pollen traits of some plum cultivars of Iran

Yavar Sharafi[1,*], Alireza Ghanbari[1, 2], Amir Mohammad Naji[3] and Allaeddin Kordenaeej[3], Ayatollah Rezaei[1]

1- Department of Horticultural Sciences, Shahed University, 1865133191, Tehran, Iran
2- Medicinal Plants Research Center, Shahed University, 1865133191, Tehran, Iran
3-Department of Agronomy and Plant Breeding, Shahed University, 1865133191, Tehran, Iran

Corresponding Author Email: y.sharafi@shahed.ac.ir

Abstract

In stone fruit trees such as plums, favorable pollination followed by good fertilization is necessary for high quality and quantity fruit yield. Therefore, pollen performance including pollen quantity produced in a flower and pollen quality consists of viability, longevity, morphological homogeneity, pollen germination and pollen tube growth rate are very important component of fertilization and fruit setting. In the study main pollen traits including germination, tube growth and longevity were investigated in ten favorable selected cultivars of plum which had been cultivated in Maragheh region, Iran. In late April of 2011, flower buds were collected in balloon stage and the pollen related parameters evaluated *in vitro* conditions immediately after sampling and also after 8 months storing in -20 °C. The results showed that there was a significant difference for pollen germination percentage and pollen tube length among the studied cultivars both immediately after sampling time and after 8 months maintenance in -20 °C. Among the studied plum cultivars, means of pollen germination percentage was ranged between 97.1-55.8% immediately after sampling time and 89.3-51.2% after 8 months maintenance in -20 °C. Also, pollen tube length among the plum cultivars were 642.4-353.6 m immediately after sampling time and 585.1-261.8 m after 8 months maintenance in -20 °C. Regarding the obtained data "Gojah Soltan" cultivar with highest pollen germination percentage and longevity could be proposed for orchard establishment and breeding programs as a pollinizer for pollination of commercially growing plum cultivars particularly in the studied area and other similar areas.

Keywords: Germination, Growth, Longevity, Plum, Pollen, Pollination

Introduction

Plum (*Prunus domestica* L.), is a temperate zone stone fruit tree which is grown in many regions with Mediterranean climates of the world. *P. cerasifera, P. domestica, P. institia* and *P. salicina* Lindl are widely grown plum species in the world. European plums *(P. domestica*) and Japanese plums (*P. salicina*) are more important in terms of commercial production (Askin and Koyuncu 1992; Faust and Surányi 1999; Karamursel et al. 2007; Son 2010). Favorable characteristics of plums are high self-fertile, effective pollen resource, easy propagation by seeds or sprouts, drought and frost resistance, good cropping, many kinds of usability for fruits, relative good resistance to fungi and parasite (Surányi 1976, 1989 and 2006). Nevertheless, European and West-Asiatic plums such as *P. insititia, P. cerasifera, P. domestica*, *P. italica* and *P. syriaca* male sterility is not been clearly known (Surányi 1976, 1989 and 2006).

Maragheh region located in northwest Iran and is a main commercially plum producing area of the country. There are about 75 native varieties of plum and prune in Iran. The most important varieties are Bokhara plum, Karaj black plum, Arak yellow plum, Baraghan prune (Goje Baraghan) and Sadie Uremia prune (Goje Sadie Uremia) (Sedaghathoor et al. 2009). The yield of plum and prune was estimated about 4 and 10 mt/ha in 2007 in the world and Iran respectively (FAOSTAT, 2009).

Some plum cultivars flower earlier in spring than most other fruit species. At this time of year the air temperature may be low and resulted in reduced pollinator activity. Some plum cultivars flower early and fruit

set can be inhibited by low temperatures at flowering time because of poor pollen germination and tube growth. Moreover, plums have strong preferences for certain pollinizers and the results are the very poor plum crops familiar to growers and most plums require cross-pollination to set commercially viable crops. Flowering and fertilization are critical for fruit set in plum trees therefore, determining the components of reproduction biology is critical for optimizing yields in orchards and so it is important for breeding programs. Some plum cultivars are self-incompatible although, hexaploid common plum *(P. domestica)* has some self-compatible cultivars (Keulemans and Van lear 1989; Keulemans 1994; Hegedus and halasz 2006; Nikolic and Milatovic 2010). Plum trees need to pollination with high quantity and quality pollens and fertilization is most important factor which affecting fruit setting. However, synchronized flowering, positive pollination and fertilization are critical for fruit set in such stated plum fruit trees (Semenas and Kouhartchik 2000; Szabo 2003).

In some breeding programs breeders maintain pollens for applying in the controlled artificial pollination methods (Parfitt and Almehdia 1984; Parfitt and Ganeshan 1989; Alburquerque et al. 2007). Artificial pollination need to using selected pollen from elite cultivars, whereas most of them are self-incompatible or have differences in blooming time and surely do not overlap each other. Due to these differences usually pollens could be collected, dried and maintained before controlled pollination programs. Meanwhile; many cultivars and genotypes with unfavorable pollens especially sterile pollens, pollens with low germination percentage and tube growth have been reported by breeders and researchers (Parfitt and Almehdia 1984; Parfitt and Ganeshan 1989; Koyuncu and Tosun 2005; Du et al. 2006; Nikolic and Milatovic 2010). Therefore, study of pollination problems and pollen traits in selected genotypes or cultivars is one of the necessary works which done in such plums specially in the commercially plum producing areas.

Many studies have investigated pollination problems and pollen viability and germination in *Prunus* species cultivars and different test methods have been used to determine the pollen viability of the fruit trees (Jefferies et al. 1982; Herrero and Arbeloa 1989; Vitagliano and Viti 1989; Cerovic 1992 and 1994). For instance, Hedly et al. (2004), Koyuncu and Tosun (2005), Du et al. (2006), Sharafi (2011a and b), and Sharafi and Bahmani (2011) investigated pollen characteristics of different stone fruit cultivars with different objectives and reported various results. Keulemans (1991 and 1994) investigated the cropping behavior, flower bud formation, pollination and fruit set of different plum cultivars in Belgium. In addition, Sedaghathoor et al. (2009) studied the morphological and pomological characteristics of 8 Iranian plum and prune cultivars in a plum orchard.

Currently it has been planned breeding programs to develop superior plum cultivars for several different usages in horticultural practices in Iran. In this regard, the study was conducted and the objective was to determine pollen longevity, viability, germination and tube growth capacity in ten favorable selected plum cultivars which are grown in different regions of Maragheh which is an important plum production area in Iran.

Materials and methods

Experimental region and plant materials

Ten favorable cultivars with high quality and quantity characteristics of the plum which called Alcha in Maragheh region (northwest of Iran) were selected for investigation. Cultivars comprised "Sardroud alcha", "Sari alcha", "Gojah soltan" ("Soltan alcha"), "Zari alcha", "Sayyd alcha", "Shalyl alcha", "Ouji biz alcha", "Ala alcha", "Eet alcha", "Gatreh tala", and "Maragheh alcha" (native to Maragheh and has very sweet fruits) which are grown in different regions of Maragheh.

Pollen bioassay

In late April of 2011, flower buds in balloon stage were collected and transmitted to laboratory. Petals and sepals were separated and anthers isolated from flower buds and placed in Petri dishes for releasing pollens. Pollens were collected and their pollen germination percentage and pollen tube length were tested immediately and then stored 8 month in -20 ^{o}C. Pollens were cultured *in vitro* medium containing 1% agar, 15% sucrose and 5 ppm boric acid, maintained about 24 hr in 24 °C and then tube growth was stopped with adding chlorophorm. Pollen germination percentage and pollen tube length were measured under light-microscope. Seven microscopic areas were counted randomly for evaluation of pollen germination percentage and pollen tube length. Pollen tube long at least as its diameter was considered to be germinated and measurements of tube length were recorded based on micrometer (µm), directly using an ocular micrometer fitted to the eyepiece on microscope.

Experimental design and data analysis

Experimental design was completely randomized design (CRD) with ten treatments (ten cultivars) with five replications (5 Petri dishes for each cultivar). Data were analyzed using SAS software and comparison of means was carried out with Duncan's multiple range tests.

Results and discussion

Analysis of variance for data showed that there were significant differences for pollen germination percentage and pollen tube length among the studied plum cultivars both immediately after sampling time and after 8 months maintenance in -20 ^{o}C (Tables 1 and 2). Among the studied plum cultivars, means of pollen germination percentage ranged between 55.8%-97.1% immediately after sampling time and 51.2%-89.3% after 8 months maintenance in -20 ^{o}C (Table 2). Maximum and minimum pollen germination percentage were observed in cultivars "Gojah soltan" (97.1%) and "Shalyl alcha" (55.8%) immediately after sampling time. However, after 8 months maintenance of pollens in -20 ^{o}C maximum and minimum germination percentage were observed in cultivars "Gojah soltan" (89.3%) and "Ouji biz alcha" (51.2%) (Table 2 and Figure 1).

Also, pollen tube length among the plum cultivars were 353.6-642.4 m immediately after sampling time and 261.8-585.1 m after 8 months maintenance in -20 ^{o}C (Table 2). Maximum and minimum pollen tube length were observed in cultivars "Gojah soltan" (642.4 m) and "Zari alcha" (353.6 m) immediately after sampling time. On the other hand, after 8 months maintenance of pollens in -20 ^{o}C maximum and minimum pollen tube length were observed in cultivars "Ala alcha" (584.1 m) and "Sardroud alcha" (261.8 m) (Table 2 and Figure 2).

According to the data it seems all of the cultivars and especially "Gojah Soltan" with highest pollen germination percentage and longevity could be selected for orchard establishment and breeding programs as a pollinizer for pollination of commercially growing plum cultivars particularly in Maragheh region of Iran.

In some stone fruit trees, pollen germination and tube growth rate are the most important characteristics related to pollen quality and also effective fertilization requires the high rates of germination and fast tube growth. Extremely low growth rates may lead to low fruit set because of ovule degradation before the pollen tube reaches to the ovary (Cheung 1996; Sharafi and Bahmani 2011). In the research, cultivars with high pollen germination percentage had not shown high pollen tube length. This phenomenon indicates genetically differences among the studied plum cultivars which have been reported previously by many researchers in almond, apricot, sweet cherry, sour cherry, plum, prune, apple, pear and other fruit trees (Stosser et al. 1996; Pirlak and bolat 1999; Sharafi 2011a and b; Sharafi and Bahmani 2011). Sometimes plum cultivars produce high quantity of pollens which does not necessarily mean high quality. Some of the pollens produced by one cultivar may be sterile or not viable (Surányi 1976, 1989 and 2006; Stosser et al. 1996; Nikolic and Milatovic 2010).

Moreover, Pirlak and Bolat (1999) by investigation on the pollen germination and pollen tube length in apricot cultivars, recorded different pollen tube lengths as 295 m in Hasanbey, 306 m in Salak, 251 m in Karacabey and 268 m in Sekerpare with 10% sucrose concentration. It is widely acknowledged that temperature and relative humidity of stored environment are the two important factors that profoundly influence the viability of stored pollen (Surányi 1976, 1989 and 2006; Stosser et al. 1996; Nikolic and Milatovic 2010).

In the study we observed some decrease in measured traits related to pollens of plum cultivars after 8 months maintenance in -20 ^{o}C in comparison with the time of their collection. Although it was observed that storage conditions below 0 °C (-20 °C and -80 °C) did not affect pollen germination after 1 year in some species of stone fruits (Parfitt and Ganeshan 1989; Parfitt and Almehdia 1989). Effect of storage temperature on the viability of pollen has been shown by some studies. For example, Alburquerque et al. (2007) studied the influence of storage temperature on the viability of pollen in 7 sweet cherry cultivars ("Brooks", "Cristobalina", "Marvin", "New Star", "Ruby" and "Somerset") and resulted that pollen viability could be maintained at reasonably high percentages after storage at -20 °C during 1 year for all studied cultivars. Also, pollen viability decreased after 15 or 30 days of storage at 4 °C. In their study, for most cultivars pollen completely lost viability after only 60 days of storage at 4°C. Remarkably, "Cristobalina" and "New Star" maintained viable pollen in relatively high percentages up to one more month at this temperature. Finally they reported that, pollen viability can be affected by long periods of storage at approximately -20 °C, being this effect genotype dependent. Similar results were also observed by Sharafi (2011 a and b) and Sharafi and Bahmani (2011), when pollen traits after short storage (6 weeks in 4°C) in some *P. domestica, P. persica, P. amygdalus* L.*, P. armeniaca* L. and *P. avium* L. genotypes were assessed.

Though there are some previous studies on the storage of pollen in some stone fruit cultivars for short or long periods of time at different temperatures, plum cultivars which studied here have been tested

for the first time in Maragheh region condition. A procedure to appropriately conserve pollen, maintaining a good viability, may allow a better planning of controlled crosses and also provide a way of exchanging pollen between breeders in different regions.

Table 1. Analysis of variances of the pollen germination percentage and pollen tube growth in studied cultivars of plum tested *in vitro* condition immediately after sampling and after 8 months maintenance in -20 °C.

Source of Variations	DF	Pollen germination (%)		Pollen tube growth (µm)	
		after sampling	after 8 months	after sampling	after 8 months
Cultivar	9	1513.7**	2059.2**	1348.1**	1319.2**
Error	40	287.9	106.3	196.2	241.3
CV (%)		11.1	15.4	13.4	12.8

**: Significant at $p \leq 0.01$ level.

Table 2. Comparison of means for pollen germination percentage and pollen tube growth in the studied cultivars of plum tested *in vitro* conditions immediately after sampling and after 8 months maintenance in -20 °C. Data are means with n = 5. In each column, mean values with different letters had significant difference at $p \leq 0.01$ according to Duncan's multiple range test.

Cultivar	Pollen germination percentage		Pollen tube growth (µm)	
	after sampling	after 8 months	after sampling	after 8 months
"Sardroud alcha"	73.2cd	65.2c	395.3ab	261.8d
"Sari alcha"	93.2a	82.1ab	435.2bc	318.7cd
"Gojah soltan"	97.1a	89.3a	642.4a	576.6a
"Zari alcha"	58.4d	56.2dc	353.6d	293.7d
"Sayyd alcha"	89.3ab	77.9b	571.9b	581.6a
"Shalyl alcha"	55.8d	54.3d	397.8c	303.5c
"Ouji biz alcha"	61.3d	51.2d	367.1d	423.1ab
"Ala alcha"	75.3c	68.2bc	588.5ab	584.1a
"Eet alcha"	84.1b	76.1b	609.7a	405.6b
"Maragheh alcha"	69.7bc	63.2c	411.7c	389.8bc

Conclusion

We concluded that pollen germination capacity and pollen tube growth rate in studied cultivars of plum were normal after 8 month maintenance in -20 °C although some decrease was observed. However based on the results, all of the investigated cultivars especially "Gojah soltan" have very vigorous pollens and they could be selected for plum orchards of Maragheh region establishment and breeding programs.

Acknowledgment

Authors would like to thank the research section of Islamic Azad University of Maragheh Branch, for their grant helps and research section of University of Maragheh for their laboratory cooperation.

References

Alburquerque N, García MF, Burgos L. 2007. Influence of storage temperature on the viability of sweet cherry pollen. Span J Agric Res 5:86–90

Askin MA, Koyuncu MA. 1992. Biological studies on some local plum cultivars, grown in Van ecological conditions. 1st Turkish National Horticulture Congress 1:17–21

Cerovic R. 1992. Pollen tube growth in sour cherry pistils in relation to fruit set. Adv Hort Sci 6:107–111

Cerovic R. 1994. Histocytological aspects of fertilization dynamics of sour cherry (*Prunus cerasus* L.). Dissertation, Faculty of Biology, Belgrade

Cheung AY. 1996. Pollen–pistil interactions during pollen tube growth. Trends Plant Sci 1: 45–51

Du YH, Zhang SL, Jiang T, Wu J (2006) Characteristics of pollen germination and pollen tube growth of *Prunus mume in vitro*. Acta Botanica 26:1846–1852.

FAOSTAT. 2009. Food and agriculture organization of the United Nations. FAO Statistics Division. http://faostat.fao.org/site/567/DesktopDefault.aspx?PageID=567

Faust M, Surányi D. 1999. Origin and dissemination of plums. Hortic Rev 23:179–23.

Hedly YA, Hormaza JI, Herrero M. 2004. Effect of temperature on pollen tube kinetics and dynamics in sweet cherry, *Prunus avium* (Rosaceae). Amer J Botany 91:558–564

Hegedus A, Halasz J. 2006. Self-incompatibility in plums (*Prunus salicina* Lindl., *Prunus cerasifera* Ehrh. and *Prunus domestica* L.). Int J Hort Sci 12:137–140

Herrero M, Arbeloa A. 1989. Influence of the pistil on pollen tube kinetics in peach (*Prunus persica*). Amer J Botany 76:1441−1447

Jefferies CJ, Brain P, Stott KG, Belcher AR. 1982. Experimental systems and mathematical models for studying temperature effects on pollen-tube growth and fertilization in plum. Plant Cell Environ 5:231−236

Karamursel OF, Sevik I, Sarisi HC, Kocal H, Ozturk FP. 2007. Adaptation of European plums (*Prunus domestica*) under Egirdir ecological conditions. 5th Turkish National Horticultur Congress 1:481−485.

Keulemans J, Van Laer H. 1989. Effective pollination period of plums: the influence of temperature on pollen germination and pollen tube growth. In: Wright CJ (ed) Manipulation of fruiting. Butterworths, London, pp 159–171

Keulemans J. 1991. Cropping behavior, flower bud formation, pollination and fruit set of different plum cultivars in Belgium. Acta Hort 283:117–129.

Keulemans J. 1994. Pollination and fruit set in self-incompatible plum cultivars. Acta Hort 359:260–268.

Koyuncu F, Tosun F. 2005. Evaluation of pollen viability and germinating capacity of some sweet cherry cultivars grown in Isparta, Turkey. 5th International Cherry Symposium. Acta Hort 1:71–75

Nikolic D, Milatovic D. 2010. Examining self-compatibility in plum (*Prunus domestica* L.) by fluorescence microscopy. Genetika 42(2):87–396

Parfitt DE, Ganeshan S. 1989. Comparison of procedures for estimating viability of *Prunus* pollen. Hort Sci 24:354–356

Parfitt DE, Almehdia E. 1984. Liquid nitrogen storage of pollen from five cultivated Prunus species. Hort Sci 19: 69–70

Pirlak L, Bolat I. 1999. An investigation on pollen viability, germination and tube growth in some stone fruits. Turk J Agric For 23:383–388

Sedaghathoor S, Ansari R, Allahyari MS, Nasiri E. 2009. Comparison of morphological characteristics of some plum and prune cultivars of Iran. Sci Res Essays 4(10): 992–996

Semenas S, Koukhartchik NV. 2000. Investigations in pollination and fertilization in *Prunus* L. breeding via fluorescence microscopy. Acta Hort 538:371–374

Sharafi Y, Bahmani A (2011) In vitro study of pollen traits after short storage in some almond, apricot and sweet cherry favorable genotypes. J Med Plant Res 5(2):266–269

Sharafi Y. 2011a. Pollen viability and longevity in some selected genotypes of peach, plum, prune and sour cherry. J Med Plant Res 5(2):275–279

Sharafi Y. 2011b. *In vitro* pollen germination in stone fruit tree of Rosaceae family. Afr J Agric Res 6(28):6021–6026

Son L. 2010. Determination on quality characteristics of some important Japanese plum (*Prunus Salicina* Lindl.) cultivars grown in Mersin-Turkey. Afr J Agric Res 5(10): 1144–1146

Stosser R, Hartman W, Anvari SF. 1996. General aspects of pollination and fertilization of pome and stone fruits. Acta Hort 423:15–21

Surányi D .2006. Comparative study of different fertile groups in plums. Int J Hort Sci 12(3):71–76

Surányi D. 1976. Differentiation of self-fertility and self-sterility in *Prunus* by stamen number/pistil length ratio. Hortscience 11(4):406–407

Surányi D. 1989. Flower structure of historical and cultivated plums, relationship between morphological characters and self fertility. Acta Bot Hung 35(1–4):199–226

Szabo Z. 2003. Plum (*Prunus domestica* L.). In: Floral biology, pollination and fertilisation in temperate zone fruit species and grape. Akadémiai Kiado, Budapest, pp 383–410

Vitagliano C, Viti R.1989. Effects of some growth substances on pollen germination and tube growth in different stone fruits. Acta Hort 239:379–381.