Research Article

Influence of Storage Duration and Storage Temperature on in-vitro Pollen Viability of Citrus Species

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Abstract

The present work was aimed to study the in- vitro pollen viability of five citrus species viz., mandarin (citrus. *reticulata* Blanco) sweet orange (*C. sinensis* L. Osbeck) grapefruit (*C. paradisi* Macf.) pummelo (*C. maxima* Burm. Merr.) and tangelo *(C. reticulata* × *c. maxima* or × *C. paradise).* The pollen at different temperatures (4ºC, -20ºC, and -196ºC) and the viability was checked from 4th up to 48 weeks. Pollen stored at low temperature (-196℃ and -20℃) showed better viability percentage as compared to pollen stored at 4℃ and room temperature. The maximum pollen viability was found in cultivar Mosambi (90.30%, 85.00%) followed by W.Murcott (86.30%, 79.00%) when stored at (-196℃ and -20℃) respectively, and it was significantly higher than all other genotypes. Minimum viability of (50.30%, 46.00%) was found in marsh seedless stored at -196℃ and -20℃ respectively. The lowest viability (16.30%) was found in marsh seedless at 4ºC at 48th week of storage interval week which was followed by 20.00% and 30.30% at -20ºC and -196ºC respectively.

At room temperature storage, the pollen viability becomes zero after the first week. The results indicate that pollen viability gradually decreased upto 48th week and pollen collected and stored at sub-zero temperatures from early blooming citrus varieties can be stored for very long period without any appreciable loss of viability and can successfully be used for throughout the blooming season for hybridization programmes by fruit breeders, for the development of new strains so as to widen the genetic base, create variability in citrus.

Keywords: Citrus, In- vitro, Pollen, Species, viability

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Introduction

Citrus is one of the world's important fruit crops which is widely grown in most areas with suitable climates between latitude 35°N~35°S. *Citrus* is the common term used for Genus, a flowering plant of *rue* family, *Rutaceae*. It falls under subfamily *Aurantioideae* and tribe *Citreae*. Although the origin of citrus is precisely not known, researchers [1-4] believe that it has the origin from south and southeast tropical and subtropical region of the world. Citrus comprises around 60 species, most of which are cultivated throughout the tropics and subtropics. They are indigenous in some parts of India, China, Northern Australia and New Caledonia [5]. Citrus in India is grown in 1.07 million ha area with a total production of 11.14 million tonnes. The most important commercial citrus groups or cultivars in India are the mandarin (C.*reticulata* Blanco) followed by sweet orange (*C. sinensis* Osbeck) and acid lime (C.*aurintifolia* Swingle) sharing 41, 23 and 23% of the respectively [6].

One of the routine tasks in plant evolutionary biology, pollination biology and crop breeding research is determining male fitness functions by quantifying total pollen grains per flower and proportion of viable pollen [7] [8]. The development of reliable methods for determining the functional quality of pollen helps in monitoring pollen vigour during storage, genetics and pollen-stigma interaction studies, crop improvement and breeding, and incompatibility and fertility studies [9]. The quality of pollen is assessed on the basis of viability and vigour of the pollen grain. Pollen vigour refers to the speed of germination of pollen grains and the rate of pollen tube growth [10]. In the breeding of fruit species it is very important to use a suitable method for determining pollen viability [11]. Several methods of pollen storage have been tried of which the most important factors are controlled temperature and humidity [12-14] and [15]. Different investigators have same consensus that low temperature and humidity are the two major influencing factors in storage of pollen grains for a long period of time [16, 17]. Pollen physiology especially germination and viability has received considerable attention for its application in plant breeding, conservation, adaptation and understanding of the physiological behaviour of the fertilizing pollen grains. There are several reports on pollen viability of different taxa [18-22] with varied aims and objectives. [23] Correlated pollen

morphology of Vitis vinifera with physiological potential by studying pollen germination in different types. This study was planned to understand the effect of low temperature storage on the viability of pollen in citrus.

Materials and Methods

Pollen from five citrus species viz. sweet orange (*c. sinensis* L. Osbeck) (Jaffa, Mosambi and Itaboria) mandarin (citrus. *reticulata* Blanco) (Daisy, W.Murcott and Kinnow) grapefruit (*c. paradisi* Macf.) (Foster, Star Ruby and Marsh Seedless) pummelo (*c. maxima* Burm. Merr.) (White Pummelo, Pink Pummelo and Devanpalli) and tangelo *(c. reticulata* × *c. maxima* or ×*c. paradise)* (Minneola and Pearl) were collected from Punjab Agricultural University Ludhiana, Punjab for the use of this study. Flowers from male parents were collected at pre blooming stage (balloon stage) and were allowed to shed the pollen in shade for 3-4 hours under 100 watt lamps. Immediately after anther dehiscence, pollen were collected in vials and subjected to different storage conditions viz. room temperature (in anhydrous calcium chloride), 4ºC, -20ºC and cryogenic storage in liquid nitrogen (-196 ºC).

Pollen viability was tested in 2 per cent acetocarmine solution, which was prepared by dissolving 2 grams of carmine powder in 45 ml of glacial acetic acid and final volume was made to 100 ml by adding distilled water. Solution was boiled for 5 minutes and filtered through Whatman No.1 filter paper. The pollen grains were dusted on a glass slide and one to two drops of acetocarmine were put on these grains. After placing a cover slip over the stain it was left for five minutes for proper staining of pollen grains. Slides were observed under microscope. Deeply stained and normal looking pollen grains were considered to be viable whereas, shrivelled, slightly stained or colourless pollen grains were counted as non viable. Three microscopic fields were observed and number of viable and non viable pollen grains was counted in each field.

$$\text{Pollen viability (\%)} = \frac{\text{No. of stained pollen}}{\text{Total no. of Pollen}} \text{ X100}$$

The experiment was carried out as completely randomized design (CRD). Data was analyzed using SAS software (SAS 9.3) and compression of means was carried out with Duncan's multiple range tests, considering $p \leq 0.05$ as the level of significance.

Results

Pollen Viability

Sweet orange (*c. sinensis* L. Osbeck) species showed 65.6% to 90.5% (**Table 1** and **Figure 1**) pollen viability of fresh pollen with maximum in Mosambi 90.5% followed by Jaffa 80.6% and minimum in Itaboria 65.6%. After 48 weeks of storage results obtained revealed that maximum average viability from 4th to 48th was noted as 78.6% in Mosambi (which was significantly higher than all other genotypes $p \leq 0.05$) at freeze temperature (-196^{0}C) and minimum as 39.1% in Itaboria at refrigerated temperature (4^{0}C). The duration starts form 4 weeks to 48 weeks i.e. for whole one year and at 4 weeks the viability was found maximum in Mosambi 90.30% at freeze temperature (-196^{0}C) and after 48 weeks of storage the viability was found 65.20% where as Itaboria at 4th weeks shows lowest viability of 50.40% at refrigerated temperature (4^{0}C) and after 48weeks viability was found 25.50% which is lowest among all sweet orange groups.

Mandarin (c. *reticulate* Blanco) species showed 80% to 86.5% (**Table 2** and **Figure 2**) pollen viability of fresh pollen with maximum in W.Murcott (86.5%) followed by kinnow (84.6) and minimum in Daisy with (80.6%). After 48 weeks of storage results obtained revealed that maximum average viability from 4th to 48th week was noted was noted as 72.8% in W.Murcott (which was significantly higher than all other genotypes $p \leq 0.05$) at freeze temperature (-196^{0}C) and minimum as 50.4% in Daisy at refrigerated temperature (4^{0}C). The duration starts form 4 weeks to 48 weeks i.e. for whole one year and at 4 weeks the viability was found maximum in W.Murcott 86.30% at freeze temperature (-196^{0}C) and after 48 weeks of storage the viability was found 57.30% where as daisy at 4 weeks shows lowest viability of 64.20% at refrigerated temperature (4^{0}C) and after 48 weeks viability was found 33.20% which is lowest among all mandarin groups.

Grapefruit (*c. paradisi* Macf.) species showed 50.6% to 65.5% (**Table 3** and **Figure 3**) pollen viability of fresh pollen with maximum in Star Ruby 65.5% followed by Foster 54.6% and minimum in Marsh Seedless 50.6%. After 48 weeks of storage results obtained revealed that maximum average viability from 4th to 48th week was noted as 53.4% in Star Ruby (which was significantly higher than all other genotypes $p \leq 0.05$) at freeze temperature (-196^{0}C) and minimum as 29.0% in Marsh Seedless at refrigerated temperature (4^{0}C). The duration starts form 4

weeks to 48 weeks i.e. for whole one year and at 4 weeks the viability was found maximum in Star Ruby 65.20% at freeze temperature (-196^{0}C) and after 48 weeks of storage the viability was found 40.40% where as Marsh Seedless at 4 weeks shows lowest viability of 38.30% at refrigerated temperature (4^{0}C) and after 48weeks viability was found 16.30% which is lowest among all Grapefruit groups.

Table 1 pollen viability % of sweet orange (*C. Sinesis* .L. Osbeck) varieties

Duration (weeks)	**Mosambi**			**Jaffa**			**Itaboria**		
	4^{o}C	**-20^{o}C**	**-196^{o}C**	**4^{o}C**	**-20^{o}C**	**-196^{o}C**	**4^{o}C**	**-20^{o}C**	**-196^{o}C**
W4	75.30	85.00	90.30	65.40	75.00	80.4	50.40	60.00	65.40
W8	72.40	82.00	87.30	62.30	72.70	78.4	47.50	58.00	61.40
W12	71.20	81.00	86.30	61.30	72.00	77.2	46.40	56.00	59.30
W16	69.20	79.00	84.30	59.40	70.00	75.4	44.30	53.00	57.50
W20	68.10	78.00	82.90	58.40	69.00	74.2	43.50	52.00	56.50
W24	66.50	76.00	81.30	56.60	67.00	71.2	41.50	50.00	54.50
W28	63.50	74.00	77.40	53.40	63.00	68.3	38.40	48.00	50.30
W32	61.30	71.00	74.20	51.50	61.00	67.3	36.30	45.00	49.30
W36	60.20	70.00	72.40	50.40	60.00	65.5	33.40	42.00	47.60
W40	58.30	67.00	71.30	47.40	59.00	64.3	31.40	40.70	46.30
W44	56.30	64.00	70.10	45.40	57.00	62.2	30.50	40.00	45.40
W48	50.40	60.00	65.20	38.30	50.00	56.3	25.50	32.00	40.50
LSD ($p \leq 0.05$)	LSD (Variety) =1.30			LSD (Variety x week) =4.50(NS)					
	LSD (Week) =2.60			LSD (Variety x Temperature) =2.30(NS)					
	LSD (Temperature) =1.30			LSD (Week x Temperature) =4.50(NS)					
				LSD (Variety x week x Temperature) =7.79					
Fresh pollen viability (%) at room temperature				80.6		90.5		65.6	
After one week pollen viability (%) at room temperature				00.00		00.00		00.00	

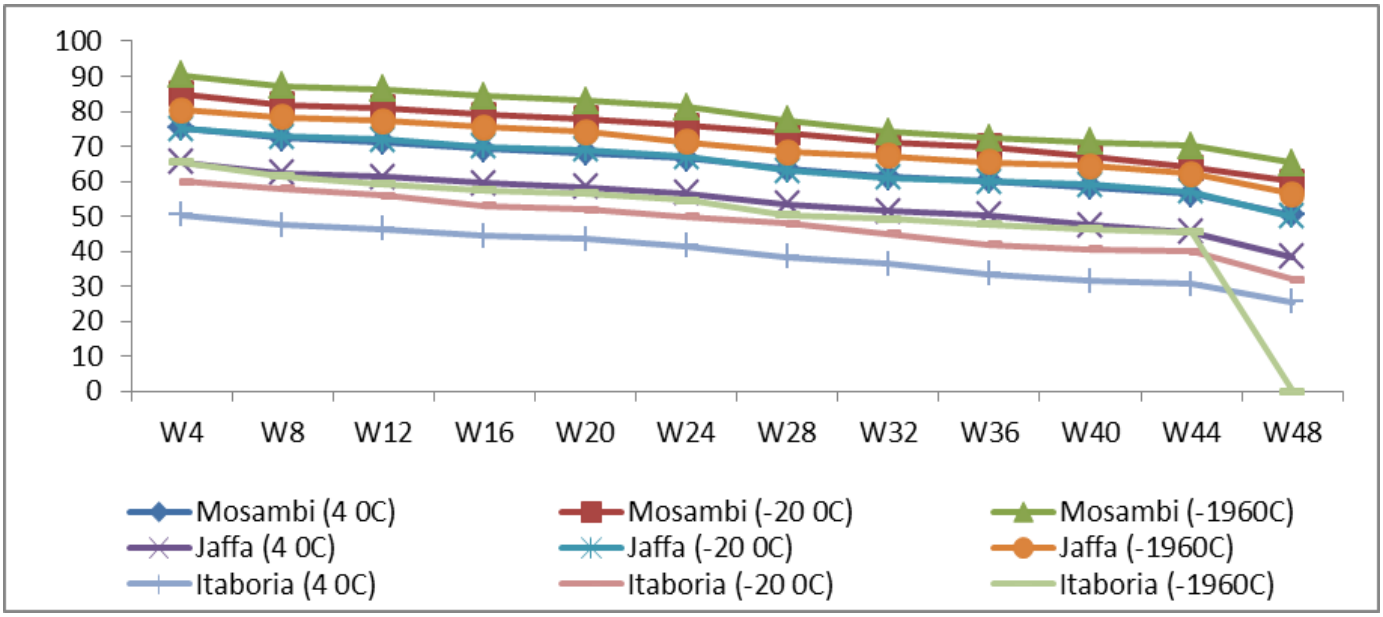


Figure 1 Pollen viability % of sweet orange (*c. sinensis* L. Osbeck) varieties

Pummelo (*c. maxima* Burm. Merr.) species showed 60.5% to 75.5% (**Table 4** and **Figure 4**) pollen viability of fresh pollen with maximum in Devanpalli 75.5% followed by White Pummelo 60.5% and minimum in Pink Pummelo 60.5%. After 48 weeks of storage results obtained revealed that maximum average viability from 4th to 48th week was noted as 65.5% in Devanpalli (which was significantly higher than all other genotypes $p \leq 0.05$) at freeze temperature (-196^{0}C) and minimum as 33.2% in Pink Pummelo at refrigerated temperature (4^{0}C). The duration starts form 4 weeks to 48 weeks i.e. for whole one year and at 4 weeks the viability was found maximum in Devanpalli 75.30% at freeze temperature (-196^{0}C) and after 48 weeks of storage the viability was found 56.30% where as Pink Pummelo at 4 weeks shows lowest viability of 45.30% at refrigerated temperature (4^{0}C) and after 48weeks viability was found 22.30% which is lowest among all Pummelo groups.

Table 2 pollen viability % of Mandarin (C. *reticulate* Blanco) varieties

Duration (weeks)	Kinnow			Daisy			W.Murcott		
	4°C	-20°C	-196°C	4°C	-20°C	-196°C	4°C	-20°C	-196°C
W4	65.30	78.00	84.40	64.20	75.00	80.30	68.30	79.00	86.30
W8	61.40	75.00	81.20	61.30	72.00	78.30	65.20	76.00	82.40
W12	60.30	73.00	80.40	60.20	71.00	77.40	64.30	74.00	80.30
W16	56.30	72.00	78.30	57.20	65.00	75.30	62.30	70.00	79.30
W20	54.40	70.00	73.20	55.20	64.00	71.30	60.20	69.00	77.30
W24	50.40	67.00	72.40	53.30	61.00	68.30	57.30	67.00	75.40
W28	48.20	64.00	70.40	50.20	60.00	66.20	55.30	65.00	70.30
W32	47.20	62.00	68.40	48.30	58.00	65.10	53.30	61.00	69.30
W36	43.30	60.00	64.30	45.30	57.00	63.20	50.30	60.00	67.40
W40	42.40	59.00	62.30	41.20	56.00	62.30	46.30	58.00	65.30
W44	40.30	57.00	60.40	38.80	53.00	60.30	44.30	56.00	63.30
W48	35.20	54.00	55.30	33.20	48.00	54.30	40.30	50.00	57.30
LSD (p≤0.05)	LSD (Variety) =1.20				LSD (Variety x week) =4.17(NS)				
	LSD (Week) =2.41				LSD (Variety x Temperature) =2.09(NS)				
	LSD (Temperature) =1.20				LSD (Week x Temperature) =4.17(NS)				
					LSD (Variety x week x Temperature) =7.22(NS)				
Fresh pollen viability (%) at room temperature					84.6		80.6		86.5
After one week pollen viability (%) at room temperature					00.00		00.00		00.00

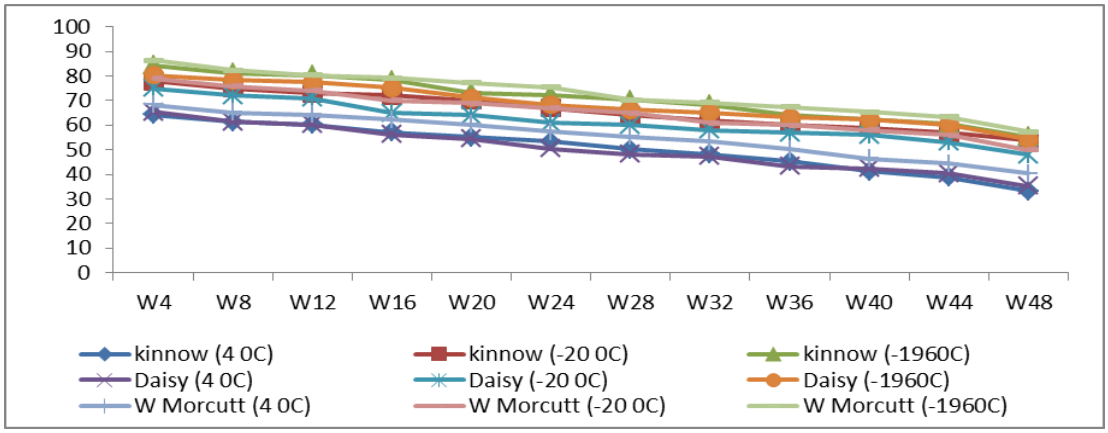


Figure 2 pollen viability % of Mandarin (c. *reticulate* Blanco) varieties

Table 3 pollen viability % of Grapefruit (*C. paradisi* Macf.) Varieties

Duration (weeks)	Foster			Star Ruby			Marsh Seedless		
	4°C	-20°C	-196°C	4°C	-20°C	-196°C	4°C	-20°C	-196°C
W4	43.30	50.00	54.40	52.40	60.00	65.20	38.30	46.00	50.30
W8	40.40	49.00	52.20	50.30	59.00	61.40	36.40	44.00	48.30
W12	39.30	47.00	51.30	49.40	57.00	60.30	35.30	43.00	47.20
W16	38.20	46.00	50.20	47.30	56.00	59.30	34.40	42.00	46.10
W20	36.40	45.00	49.20	45.30	53.00	56.40	32.40	40.00	43.40
W24	35.30	43.00	48.10	42.30	50.00	53.30	30.30	38.00	41.30
W28	30.40	41.00	45.40	41.50	49.00	52.30	28.30	36.00	40.30
W32	27.40	40.00	42.40	37.40	47.00	50.40	27.30	35.00	37.30
W36	25.30	38.00	41.20	36.30	46.00	49.50	25.40	33.00	36.40
W40	20.40	37.00	39.20	35.30	44.00	47.50	23.40	32.00	35.50
W44	18.40	36.00	38.30	32.40	41.00	45.30	20.30	30.00	33.30
W48	12.40	30.00	34.30	26.40	34.00	40.40	16.30	20.00	30.30
LSD (p≤0.05)	LSD (Variety) =1.19				LSD (Variety x week) =4.13(NS)				
	LSD (Week) =2.38				LSD (Variety x Temperature) =2.06				
	LSD (Temperature) =1.19				LSD (Week x Temperature) =4.13 (NS)				
					LSD (Variety x week x Temperature) =7.15				
Fresh pollen viability (%) at room temperature					54.6		65.5		50.6
After one week pollen viability (%) at room temperature					00.00		00.00		00.00

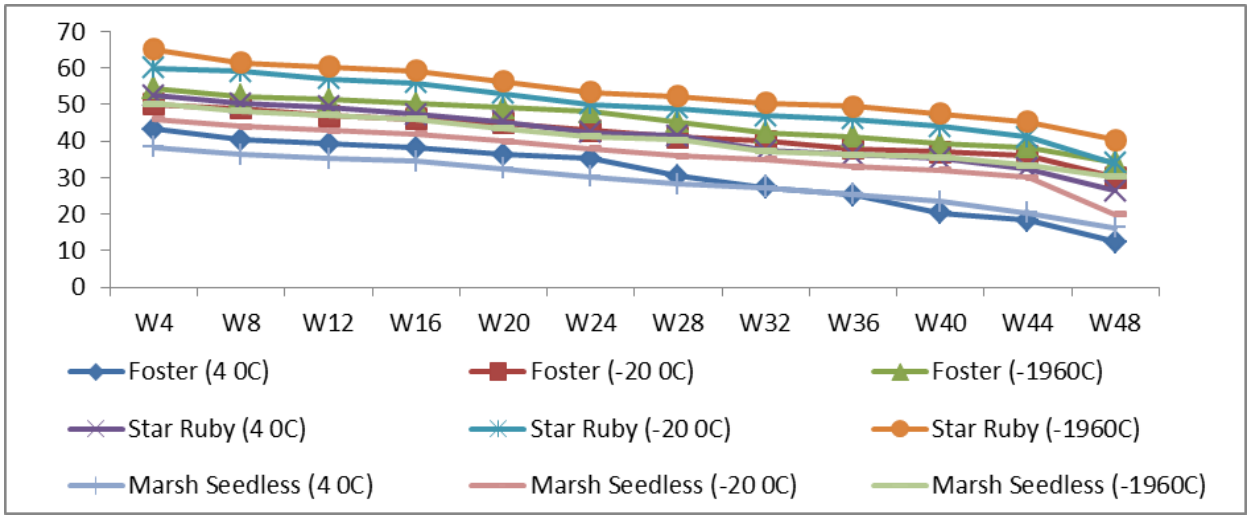


Figure 3 pollen viability % of Grapefruit (*c. paradisi* Macf.) Varieties

Table 4 pollen germination % of Pummelo (*C. maxima* Burm. Merr.) Varieties

Duration (weeks)	Pink Pumello			White Pumello			Devanpalli		
	4°C	-20°C	-196°C	4°C	-20°C	-196°C	4°C	-20°C	-196°C
W4	45.30	55.00	60.40	52.20	60.00	65.30	67.30	72.00	75.30
W8	43.60	52.00	57.30	50.30	57.00	62.40	66.30	70.00	73.30
W12	42.30	51.00	56.50	49.20	56.00	60.60	64.30	67.00	71.30
W16	40.50	49.00	54.40	47.30	54.00	59.40	63.40	65.00	70.20
W20	39.10	48.00	53.20	45.00	53.00	58.40	61.50	64.00	69.30
W24	37.20	46.00	51.20	44.30	51.00	56.50	59.40	62.00	67.40
W28	32.40	41.00	46.30	40.20	48.00	53.30	57.40	60.00	64.30
W32	28.40	38.00	44.30	38.30	45.00	51.40	56.50	59.00	63.30
W36	25.30	36.00	40.40	35.30	43.00	49.40	54.20	57.00	61.30
W40	24.40	34.00	38.40	34.40	41.00	47.40	53.50	56.00	58.30
W44	22.30	31.00	35.30	32.30	40.00	46.50	50.40	55.00	56.30
W48									
LSD (p≤0.05)	LSD (Variety) =1.14			LSD (Variety x week) =3.96 (NS)					
	LSD (Week) =2.28			LSD (Variety x Temperature) =1.98					
	LSD (Temperature) =1.14			LSD (Week x Temperature) =3.96(NS)					
				LSD (Variety x week x Temperature) =6.85					
Fresh pollen viability (%) at room temperature				60.5		65.6		75.5	
After one week pollen viability (%) at room temperature				00.00		00.00		00.00	

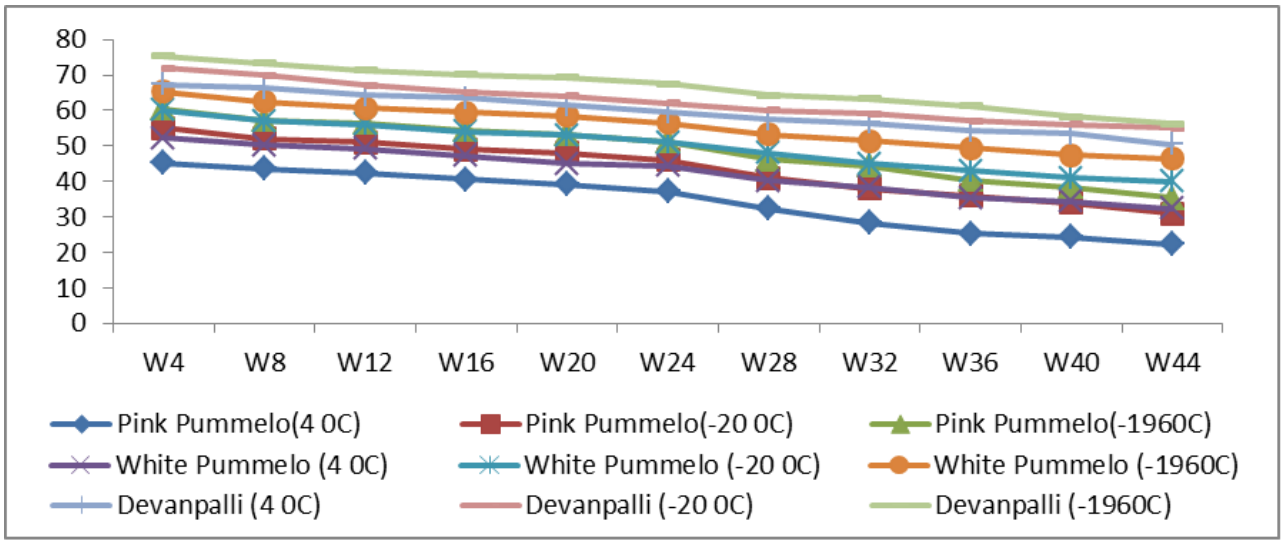


Figure 4 pollen germination % of Pummelo (*c. maxima* Burm. Merr.) Varieties

Table 5 Pollen viability % of Tangelo *(c. reticulata × c. maxima* or *× c. Paradise)* varieties

Duration (weeks)	Minneola			Pearl		
	4°C	-20°C	-196°C	4°C	-20°C	-196°C
W4	54.30	70.00	76.20	60.00	78.00	85.00
W8	51.00	68.00	73.40	57.00	75.00	83.00
W12	50.40	65.00	71.30	53.00	72.00	81.00
W16	47.30	63.00	69.50	51.00	71.00	80.00
W20	43.20	60.00	63.30	49.00	69.00	78.00
W24	41.40	58.00	60.30	46.00	66.00	76.00
W28	40.50	55.00	57.60	44.00	63.00	73.00
W32	38.40	53.00	55.30	42.70	60.00	70.00
W36	36.40	50.70	53.40	41.00	58.70	68.00
W40	33.30	50.00	50.40	39.00	57.00	64.00
W44	31.40	48.00	48.40	37.00	55.00	62.00
W48	25.40	40.00	40.30	32.00	50.00	60.00
LSD (p≤0.05)	LSD (Variety) =1.14			LSD (Variety x week) =3.96(NS)		
	LSD (Week) =2.80			LSD (Variety x Temperature) =1.98		
	LSD (Temperature) =1.40			LSD (Week x Temperature) =4.85(NS)		
				LSD (Variety x week x Temperature) =6.86(NS)		
Fresh pollen viability (%) at room temperature				76.6	85.6	
After one week pollen viability (%) at room temperature				00.00	00.00	

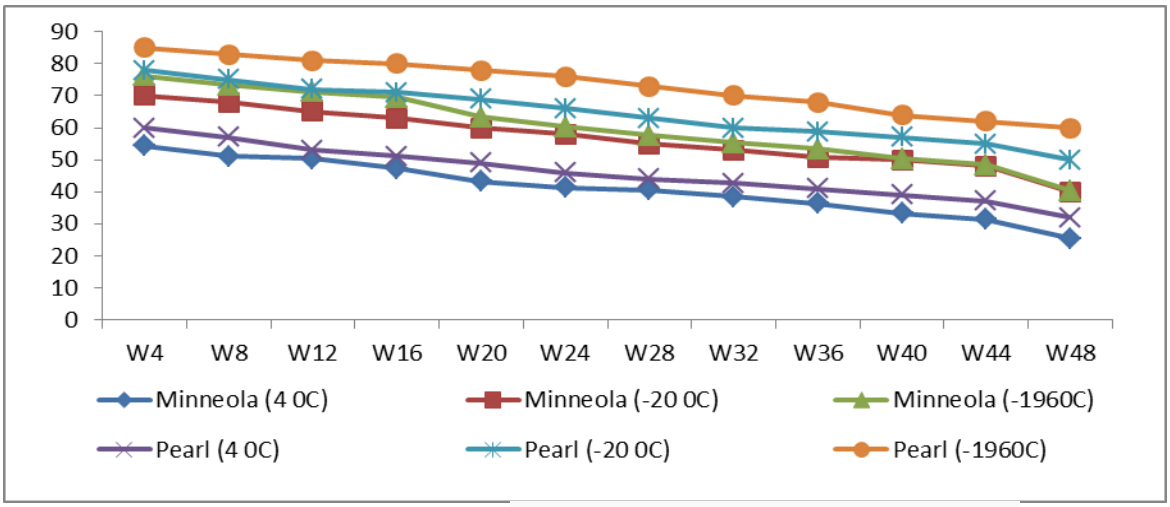


Figure 5 Pollen viability % of Tangelo *(c. reticulata × c. maxima* or *× c. Paradise)* varieties

Table 6 Combined interaction of all varieties, weeks and temperature among each other and their values

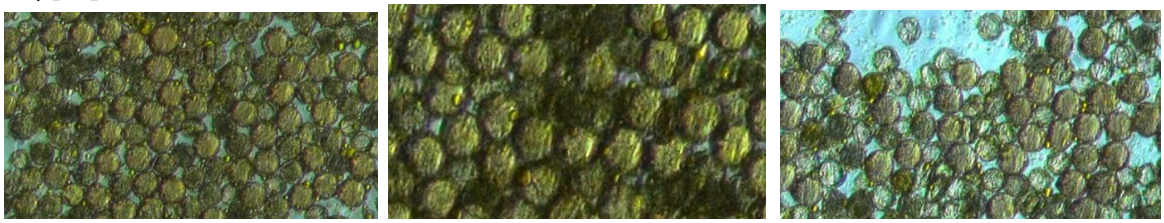
LSD (p≤0.05)	LSD (Variety)	=1.20	LSD (Variety x week)	=4.14(NS)
	LSD (Week)	=1.11	LSD (Variety x Temperature)	=2.07
	LSD (Temperature)	=0.55	LSD (Week x Temperature)	=1.92(NS)
			LSD (Variety x week x Temperature)	=7.17

Tangelo (c. reticulata × c. maxima or ×c. paradise) species showed 76.6% to 85.6% (**Table 5** and **Figure 5**) pollen viability of fresh pollen with maximum in Pearl 85.6% and minimum in Minneola 76.6%. After 48 weeks of storage results obtained revealed that maximum average viability from 4th to 48th week was noted as 73.3% in Pearl (which was significantly higher than all other genotypes $p \leq 0.05$) at freeze temperature (-196^{0}C) and minimum as 41.1% in Minneola at refrigerated temperature (4^{0}C). The duration starts form 4 weeks to 48 weeks i.e. for whole one year and at 4 weeks the viability was found maximum in Pearl 85.00% at freeze temperature (-196^{0}C) and after 48 weeks of storage the viability was found 60.00% where as Minneola at 4 weeks shows lowest viability of 54.30% at refrigerated temperature (4^{0}C) and after 48 weeks viability was found 25.40% which is lowest among all Tangelo groups.

Discussion

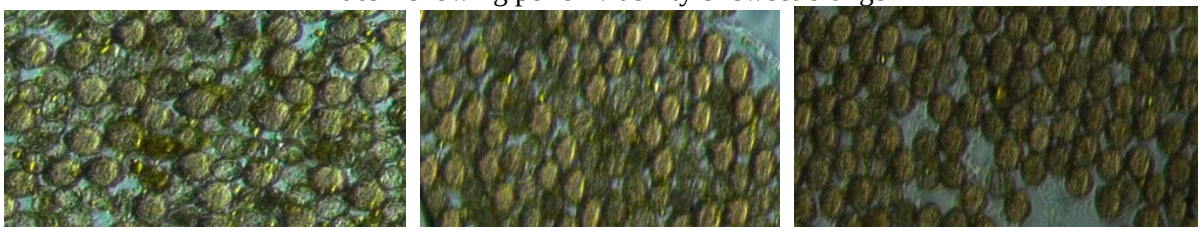
Pollen viability was examined up to 48 weeks in different storage conditions viz. 4 ^{0}C, -20 ^{0}C and -196 ^{0}C (Table 1 and Figure 1). The results obtained revealed that pollen viability was more than 50.00 per cent in all the fourteen cultivars immediately after gathering in laboratory with maximum in Mosambi (90.5%) (which was significantly higher than all other genotypes $p \leq 0.05$) followed by W.Murcott (86.5%) and minimum in Marsh Seedless (50.6 %). While comparing different storage temperatures, the viability was lost immediately after storage in case of room temperature. However, maximum viability was observed at sub zero storage temperatures (-196^{0}C) during the whole period of storage duration with highest average pollen viability was noted in cultivar Mosambi (78.6 %) (**Table 6**) at freeze temperature (-196^{0}C) from 4th to 48th week (which was significantly higher than all other genotypes $p \leq 0.05$) and lowest in Marsh seedless (40.8%) at refrigerated temperature (4^{0}C). Minimum viability was observed of (38.30%) in cultivar Marsh Seedless at refrigerated temperature (4^{0}C) at 4th week and after 48th week it and lowest of (16.30%). The viability showed a decreasing trend with increase in storage period and thus an inverse relation between viability and duration of storage was observed. Similar finding was done by [24-27]. [28] concluded that pollen of lemon (cv. Meyer) had the highest viability with 86.74%. A decreasing trend in viability was observed with increase in storage period at all the storage temperatures however minimum loss was observed at – 196 ^{0}C (78.6% to 40.8%), (which was significantly higher than all other genotypes $p \leq 0.05$) followed by -20 ^{0}C (73.9% to 36.6%) and 4^{0}C (64.4% to 29.0%) respectively. Thus variability among different cultivar was observed for their viability potentials.

In the present study wide variations in viability of pollen grains was observed with different storage temperatures and durations among different citrus cultivars. This variability may be due to pollen fertility, as a result of regular meiosis and activation of certain enzyme systems present in the pollen grain itself. Besides genotype and environmental interactions may also play an important role. This phenomenon indicates genetic differences among the genotypes which have been reported by many researchers in many of the fruit tree species and cultivars [29, 30]. [31] five *Citrus* species while studying pollen germination beyond 48 weeks in the refrigerator (4^{0}C), freezer (-20^{0}C, -30^{0}C) and freeze drier (-60^{0}C) the best method to maintain pollen seems to be freeze drier (-60^{0}C) and the viability of stored pollen grains for a long period of time. Among five species *C.* aurantium, *C.* limon and *C.* sinensis showed high percentage of germination as compared to *C.* reticulata and *C.* paradisii. The gradual loss of germination at low temperatures (-20^{0}C and -196^{0}C) observed in the present study may be attributed to frequent freezing and thawing of pollen grains. Furthermore low temperature might have lead to intracellular ice formation, cell death and thereby loss of germination. Our results clearly indicated that it is feasible to store pollen grains of citrus at sub zero temperatures without any significant loss in their viability and germinability, and they may be used effectively throughout the flowering season for assisted pollination so as to broaden the citrus genetic base. Similarly in case of pollen germination the cultivar Mosambi had maximum average germination percentage when pollens were stored at -196^{0}C (70.8 %,) -20^{0}C (66.3 %,) which was significantly higher than all other genotypes and followed by W.Murcott - 196^{0}C (60.9%) -20^{0}C (48.5%). Minimum germination percentage was found in Marsh Seedless stored at -196^{0}C (21.7%) -20^{0}C (12.5%) [32].



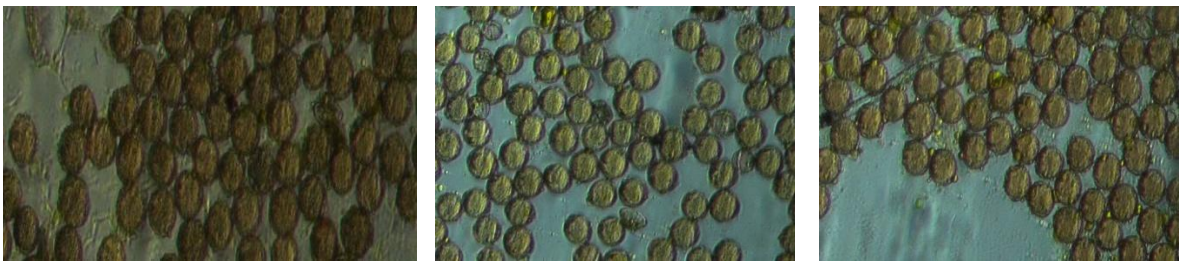
Mosambi Jaffa Itaboria

Plate 1 showing pollen viability of sweet orange



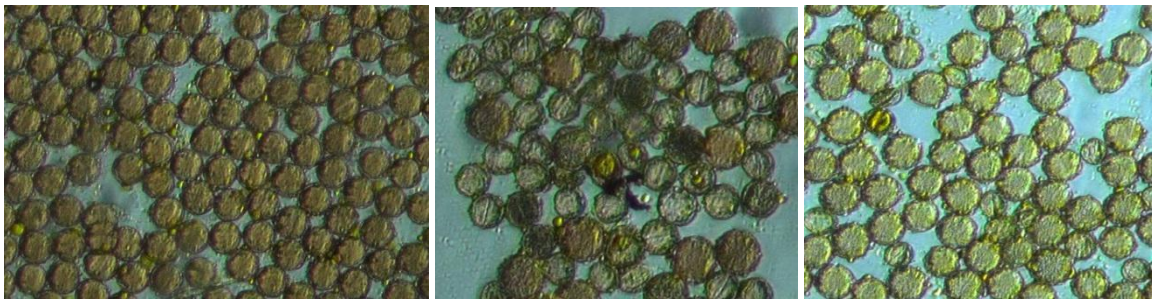
W. Murcott Kinnow Daisy

Plate 2 showing the pollen viability of mandarin



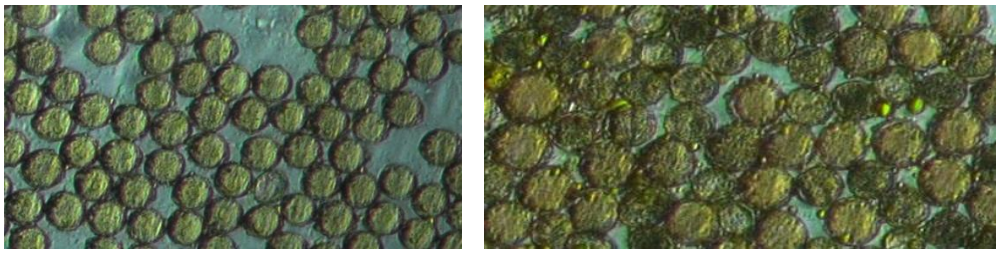
Star Ruby Marsh Seedless Foster

Plate 3 showing pollen viability of grapefruit



Devanpalli White Pummelo Pink Pummelo

Plate 4 showing the pollen viability of pummelo



Minneola Pearl

Plate 5 Plate showing pollen viability of tangelo

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