Influence of different varieties, media and storage temperature on pollen viability in mango (Mangifera indica L.)

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Abstract
Study was conducted to investigate pollen viability of four mango varieties, viz. Kesar, Alphonso, Mallika and Dudhpendo up to 56 days. Pollen grains were suspended in three different media (n-hexane, paraffin oil and without media) under four different storage temperature (20 °C, -4 °C, 4 °C and room temperature). Aceto-carmine test was used to determine pollen viability. Variation due to different varieties, media, temperature and their interactions was found significant. Among four varieties, maximum viability was recorded in Kesar during 7, 14, 21, 49 and 56 days of storage, whereas, in Mallika, at 28, 35 and 42 days. In storage media, n-hexane was found best in terms of retaining viability. The storage at -20 °C gave better results in terms of viability in all four varieties. On the basis of results, it could be concluded that pollen grains of Kesar and Mallika could be stored successfully up to 56 days in n-hexane at -20°C.

Keywords: Mango, pollen viability, aceto-carmine test, storage media, storage temperature

Introduction
Mango is one of the major fruit crops of Asia and has developed its own importance all over the world. It occupies a unique place among all the fruits grown in India. Mango (2n=40), is an allopolyploid, most probably amphidiploids and cross pollinated species. Pollination in mango is a complex physiological phenomenon. Both self and cross pollination have been documented in mango; but unreliable results have been reported on cross-pollination mechanisms in most parts of the world. Mango productivity is badly affected by irregular flowering, proportion of bisexual flowers, poor pollination and fertilization and fruit drop. Flowering times of mango vary greatly among different cultivars and environment. It ranged from December to April depending on different agro-climatic zones of the country. Thus, in order to optimize the pollination for both early and late blooming cultivars, it would be necessary to conserve pollen from early blooming cultivars for pollination in late season in late blooming cultivars and vice versa. Mango pollen grain is characterized by short viability and high sensitivity to desiccation, consequently, its conservation is very problematic (Issarakraisila and Considine, 1994) [13]. Pollen viability and its germination are also cultivar dependent characters in mango. (Abourayya et al., 2011; Singh, 1954 and Desai et al., 1986) [1, 23, 10]. Mango pollen requires some specific conditions of temperature and humidity as well as an artificial growth medium to achieve germination equaling that of pollen on stigmas in vivo (De Wet and Robbertse, 1986) [9]. Pollen storage is used to extend its longevity and allows its use in pollination performed at a later date, thus overcoming crossing barriers due to asynchronous flowering between individuals in mango. Short-term storage of pollen provides a breeder with viable pollen within a flowering season and allows pollination of a late emerging flower with an earlier flowering genotype (Chaudhury et al., 2010; Dutta et al., 2013 and Mishra and Shivanna, 1982) [7, 11, 18]. To fulfill the following objective, an attempt was made to determine the pollen viability of different mango cultivars under different storage media and to standardise the storage temperature for mango pollen storage.

Materials and Methods
Four mango varieties viz., Kesar, Alphanso, Dudhpendo and Mallika were chosen for present study available at Fruit Research Station, Sakkarbaug, Junagadh Agricultural University Junagadh.

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The pollen used in this experiment was collected from freshly opened flowers of all four mango cultivars between 8:00 and 10:00 am (±23 °C). Flowers were collected prior to anthesis, a stage characterized by pink anther colour. The harvested flowers were placed in the sun to prompt anther dehiscence. Freshly dehisced anthers, with pollen grains visible on the anthers as a greyish powder, were collected in a petri dish using forceps and dried in a desiccator for two hours. Dried pollen grains were suspended in the different three storage media, viz. n-hexane, paraffin oil and without media and stored at room temperature in a controlled growth room (27 °C) and at low temperatures in a cold-storage facility (4 °C, -4 °C, -20 °C) at the Post Graduate Lab, College of Horticulture, Junagadh.

The samples of pollen grains were taken from storage each week for observations. Pollen grains were separated from oil/organic solvent via a filtration process. Filter paper was also used in oil treatments to remove the paraffin oil from the pollen surface. Paraffin oil along with pollen grains was poured onto the filter paper directly from the petri dishes and allowed to filter through, leaving pollen grains on the paper. The pollen grains on the filter paper were then rinsed 4–5 times with n-hexane to remove residual oil, before they were dried for 10–15 minutes. Pollen viability was estimated using aceto-carmine test. This method includes the addition of colorant on pollen. The pollen nucleus is rich in chromatin material and viable pollen stains pink to deep red with aceto-carmine, while sterile (mostly shrivelled) pollen does not take any stain and thus remains pink to deep red with aceto-carmine, while sterile (mostly shrivelled) pollen does not take any stain and thus remains pink to deep red with aceto-carmine, while sterile (mostly shrivelled)

Results and Discussion

Effect of variety on mango pollen viability

In case of varieties, the variation was found significant for all storage days (Table 1). Significantly maximum pollen viability (63.67, 50.36, 38.56, 7.74 and 3.74%) were recorded in Kesar (V₁) which was at par with Mallika (V₃) during 7, 14, 21, 49 and 56 days of storage, respectively. Whereas, maximum pollen viability (27.99, 21.26 and 14.41%) were noted in Mallika (V₃) at 28, 35 and 42 days of storage. Likewise, minimum pollen viability was recorded in Alphonso (V₂) during all days of storage, respectively.

The variation in pollen viability among the various varieties might be due to the genetic constitution of varieties and their interaction with climatic parameters. This phenomenon indicated genetic differences among the genotypes which have been reported by many researchers in many of the fruit tree species and cultivars. (Alburquerque et al. 2007; Sharafi et al. 2011) [3, 21]. Kesar and Mallika had a good pollen viability might be due to its more tolerability against climatic parameters.

During all storage days, lowest pollen viability was observed in variety Alphonso (V₂). According to Shivashankara et al. (2019) [22], Alphonso pollens had higher amino acids and sugars at anthesis later on they decreased rapidly. This indicates that these pollen grains may be lacking the enzymes which are useful for the utilisation of sugars and amino acids for germination. So that the pollen grains of Alphonso cultivar is degraded rapidly compared to other cultivars. The result was in confirming those by Abourayya et al. (2011) [11], Dutta et al. (2013) [13] and Shivashankara et al. (2019) [22] in mangoes. Present finding were also in agreement with Salles et al. (2007) [20], Bhat et al. (2012) [8], Coser et al. (2012) [18], Gaaliche et al. (2013) [12], Soares et al. (2016) [24], Ahmed et al. (2017) [17] and Mesnoua et al. (2018) [18] in other fruit crops.

Table 1: Effect of different mango variety, media and temperature on pollen viability at 35, 42 and 56 days of storage

| Treatments                  | Pollen viability (%) |
|-----------------------------|----------------------|
|                            | 7        | 14       | 21       | 28       | 35       | 42       | 49       | 56       |
| Factor A (Variety - 4)      |          |          |          |          |          |          |          |          |
| V₁: Kesar                   | 63.67    | 50.36    | 38.56    | 27.85    | 20.21    | 14.26    | 7.74     | 3.74     |
| V₂: Alphonso                | 56.43    | 44.97    | 31.14    | 21.28    | 15.20    | 9.76     | 3.97     | 0.91     |
| V₃: Mallika                 | 62.82    | 48.43    | 38.15    | 27.99    | 21.26    | 14.41    | 7.24     | 3.26     |
| V₄: Dudhpendo               | 60.75    | 45.99    | 33.94    | 25.66    | 17.50    | 11.37    | 6.63     | 2.10     |
| S.Em. ±                     | 0.25     | 0.27     | 0.31     | 0.27     | 0.27     | 0.18     | 0.14     | 0.05     |
| C.D. at 5%                  | 0.71     | 0.75     | 0.86     | 0.75     | 0.75     | 0.50     | 0.38     | 0.13     |
| Factor B (Media - 3)        |          |          |          |          |          |          |          |          |
| M₁: n-hexane                | 73.63    | 61.03    | 49.63    | 39.73    | 32.23    | 23.21    | 13.19    | 5.57     |
| M₂: Paraffin oil            | 64.51    | 52.99    | 42.80    | 32.11    | 23.39    | 14.15    | 6.00     | 1.94     |
| M₃: No media (Control)      | 44.61    | 28.29    | 13.90    | 5.25     | 0.00     | 0.00     | 0.00     | 0.00     |
| S.Em. ±                     | 0.22     | 0.23     | 0.27     | 0.23     | 0.23     | 0.15     | 0.12     | 0.04     |
| C.D. at 5%                  | 0.62     | 0.65     | 0.74     | 0.65     | 0.65     | 0.43     | 0.33     | 0.12     |
| Factor C (Storage Temperature - 4) |          |          |          |          |          |          |          |          |
| S₁: -20 °C                  | 81.67    | 69.15    | 55.70    | 41.98    | 31.22    | 23.38    | 15.41    | 8.95     |
| S₂: -4 °C                   | 73.97    | 60.17    | 46.29    | 33.10    | 22.93    | 14.01    | 7.45     | 1.06     |
| S₃: +4 °C                   | 64.36    | 51.09    | 38.22    | 27.70    | 20.01    | 12.42    | 2.72     | 0.00     |
| S₄: Room temperature       | 23.67    | 9.34     | 1.57     | 0.00     | 0.00     | 0.00     | 0.00     | 0.00     |
| S.Em. ±                     | 0.25     | 0.27     | 0.31     | 0.27     | 0.27     | 0.18     | 0.14     | 0.05     |
| C.D. at 5%                  | 0.71     | 0.75     | 0.86     | 0.75     | 0.75     | 0.50     | 0.38     | 0.13     |

Interactions

| V × M                      |          |          |          |          |          |          |          |          |
| S.Em. ±                    | 0.44     | 0.46     | 0.53     | 0.46     | 0.46     | 0.31     | 0.23     | 0.08     |
| C.D. at 5%                 | 1.23     | 1.29     | 1.30     | 1.30     | 0.87     | 0.66     | 0.23     |          |
| V × S                      |          |          |          |          |          |          |          |          |
| S.Em. ±                    | 0.51     | 0.53     | 0.61     | 0.53     | 0.53     | 0.36     | 0.27     | 0.10     |
| C.D. at 5%                 | 1.42     | 1.49     | 1.72     | 1.50     | 1.50     | 1.00     | 0.76     | 0.27     |
| M × S                      |          |          |          |          |          |          |          |          |
| S.Em. ±                    | 0.44     | 0.46     | 0.53     | 0.46     | 0.46     | 0.31     | 0.23     | 0.08     |
| C.D. at 5%                 | 1.23     | 1.29     | 1.49     | 1.30     | 1.30     | 0.87     | 0.66     | 0.23     |
Effect of media on mango pollen viability

The variation observed in pollen viability due to storage media was also found significant for all storage days (Table-1). Highest pollen viability (73.63, 61.03, 49.63, 39.73, 32.23, 23.21, 13.19 and 5.57%) were registered in pollen grains stored with media n-hexane (M₁) followed by paraffin oil (M₂) during all storage days, respectively. There was no viability found in pollen gains stored in control condition without any media (M₃) after 28 days of storage.

The variation presented in pollen viability might be due to the sensitivity of mango pollen to extended storage media exposure. However, solvent medium has proved to be better for the storage of any pollen at low temperature (Mishra and Shivanna, 1982) [18]. In non-polar solvents such as cyclohexane, hexane and diethyl ether, pollen grains could be stored successfully due to very little leaching of membrane phospholipids, sugars and amino acids into the solvent. In polar solvents, however, extensive leaching of these compounds leads to loss of pollen viability. Results were also in close agreements with the finding of Kumar et al. (2015) [16] as well as with Jain and Shivanna (1988, 1990) [14].

Effect of storage temperature on mango pollen viability

It has been widely acknowledged that temperature and relative humidity of the storage environment are two important factors, which profoundly influence the viability of stored pollen. The variation in pollen viability due to different storage temperature was also observed significant (Table-1). Maximum pollen viability (81.67, 69.15, 55.70, 41.98, 31.22, 23.38, 15.41 and 8.95%) were noted in pollen grains stored at temperature -20 °C (S₁) followed by -4 °C (S₂) during all days of storage, respectively. The differential rate of reduction in pollen viability stored at different temperature regimes may be because of rate of metabolic activities in pollen in temperature dependent. At 28 days of storage, pollen grain stored at room temperature was completely lost its viability. The pollen viability decreased most rapidly at room temperature, mainly due to the high storage temperature, strong activity in pollen, accelerated metabolism, excessive consumption of nutrients stored in pollen. The result was confirming with Dutta et al. (2013) [11] who concluded that room temperature storage of Sensation, Tommy Atkins and Janardan Pasand showed only 2.11%, 1.80% and 1.03% pollen viability after 4 weeks (28 days) of storage where they basically became non-viable. Pollen storage of mango cultivars at -20 °C showed the percentage of viability was high in all three mango cultivars at all dates of observations compared to pollen storage at -4 °C. Present results were also in agreement with Alburquerque et al. (2007) [13], Salles et al. (2007) [20], Thaipong et al. (2008) [23], Mesnoua et al. (2018) [17] and Chandler et al. (2019) [26] in sweet cherry, citrus, grape, date palm and sugar apple, respectively. Our results clearly indicated that it is feasible to store pollen grains of mango at sub-zero temperatures (-20°C) without any significant loss in their viability which was confirming with Bhat et al. (2012) [5] in pear.

Interaction effect of variety and media on mango pollen viability

The interaction effect of different variety and media on pollen viability was found significant at all days of storage except 21 days (Table-2). Maximum pollen viability (77.88, 65.39, 15.97 & 8.03%) were noted in variety Kesar stored with media n-hexane (V₃M₁), respectively followed by V₃M₄ at 7, 14, 49 and 56 days of storage. Likewise, maximum pollen viability (42.87, 37.30 & 27.68%) was noted in variety Mallika stored with media n-hexane (V₄M₁) at 28, 35 and 42 days of storage, respectively. Pollen grains of all four varieties stored without media had no viability from 35 days of storage. The variation in pollen viability among the various varieties might be due to the genetic constitution of varieties and their positive interaction with storage media. Kesar and Mallika had good pollen viability in storage media n-hexane could be due to very little leaching of membrane phospholipids, sugars and amino acids into the solvent and their good genetic makeup compared to others.

| V × M × S | S.Em. ± | C.D. at 5% | C.V. % |
|-----------|---------|-----------|-------|
| 0.88      | 0.92    | 1.06      | 0.92  |
| 2.47      | 2.59    | 2.98      | 2.59  |
| 2.50      | 3.37    | 5.18      | 6.22  |
|           |         |           | 8.63  |
|           |         |           | 8.59  |
|           |         |           | 12.71 |
|           |         |           | 11.46 |

Table 2: Interaction effect of variety and media on pollen viability at all days of storage except 21 days
Interaction effect of media and storage temperature on mango pollen viability

Variation due to interaction effect of different media and storage temperatures on pollen viability was also found significant at all days of storage (Table-3). Maximum pollen viability (93.42, 84.89, 72.84, 60.18, 53.87, 42.58, 29.93 & 19.08%) were noted in pollen grains stored with n-hexane at -20°C (M₁S₁) at all days of storage, respectively. Minimum pollen viability (12.17 & 0.80%) were noticed in pollen grains stored without media at room temperature (M₃S₄) at 7 and 14 days of storage, respectively which turns to zero from 21 days of storage. Pollen viability also reduced to zero in all samples stored at room temperature with n-hexane as well as paraffin oil from 35 days of storage. As mango pollen was more delicate to desiccation and high temperature, pollen viability was decreased at room temperature after 14 days of storage without any media.

Table 3: Interaction effect of media and storage temperature on pollen viability

| Treatments M x S | Pollen viability (%) |
|------------------|----------------------|
| M₁ S₁           | 93.42 84.89 72.84 60.18 53.87 42.58 29.93 19.08 |
| M₁ S₂           | 86.12 75.06 63.98 51.26 38.66 25.58 14.66 3.16 |
| M₁ S₃           | 79.01 68.92 58.35 47.46 36.38 24.66 18.16 0.00 |
| M₁ S₄           | 35.97 15.27 3.37 0.00 0.00 0.00 0.00 0.00 |
| M₂ S₁           | 84.37 73.15 66.12 51.79 39.79 27.55 16.30 7.77 |
| M₂ S₂           | 79.09 68.24 55.15 41.02 30.13 16.46 7.71 0.00 |
| M₂ S₃           | 71.73 58.63 48.59 35.64 23.65 12.59 0.00 0.00 |
| M₂ S₄           | 22.86 11.94 1.35 0.00 0.00 0.00 0.00 0.00 |
| M₃ S₁           | 67.23 49.41 28.15 13.98 0.00 0.00 0.00 0.00 |
| M₃ S₂           | 56.69 37.22 19.73 7.02 0.00 0.00 0.00 0.00 |
| M₃ S₃           | 42.35 25.72 7.72 0.00 0.00 0.00 0.00 0.00 |
| M₃ S₄           | 12.17 0.80 0.00 0.00 0.00 0.00 0.00 0.00 |
| S.Em. ±         | 0.44 0.46 0.53 0.46 0.46 0.31 0.23 0.08 |
| C.D. at 5%       | 1.23 1.29 1.49 1.30 1.30 0.87 0.66 0.23 |
| C.V. %           | 2.50 3.37 5.18 6.22 8.63 8.59 12.71 11.46 |

Interaction effect of variety and storage temperature on mango pollen viability

Similarly, variation due to the interaction effect of different variety and storage temperature on pollen viability was also found significant (Table-4). Maximum pollen viability (84.16, 70.18, 55.45 & 26.80%) were noted in Mallika stored at -20°C (V₁S₁) at 7, 14, 35 and 42 days of storage, respectively. At 21, 28, 49 and 56 days of storage, maximum pollen viability (59.67, 45.89, 17.52 & 12.76%) were noted in variety Kesar stored at -20°C (V₁S₁), respectively. Minimum pollen viability was noted in variety Alphonso stored at room temperature (V₂S₄) at 7 and 14 days of storage but from 21 days of storage, it turns to zero viability in all varieties. The decrease in pollen viability at room temperature might be attributed to high sensitivity of mango pollen to existing high temperatures and low relative humidity. The result was confirming with Alburquerque et al. (2007) [3], Bhat et al. (2012) [8], Dutta et al. (2013) [11] and Bath et al. (2017) [4] in sweet cherry, pear, mango and citrus, respectively.

Table 4: Interaction effect of variety and storage temperature on pollen viability

| Treatments V x S | Pollen viability (%) |
|------------------|----------------------|
| V₁ S₁           | 81.51 70.03 59.67 45.89 34.49 26.32 17.52 12.76 |
| V₁ S₂           | 73.49 62.60 47.49 35.24 24.78 15.93 9.90 2.21 |
| V₁ S₃           | 69.91 55.99 42.35 30.27 21.56 14.77 3.55 0.00 |
| V₁ S₄           | 29.79 12.80 4.72 0.00 0.00 0.00 0.00 0.00 |
| V₂ S₁           | 78.65 67.32 52.63 37.85 25.92 17.75 10.12 3.63 |
| V₂ S₂           | 69.58 57.42 41.92 25.83 20.02 12.47 5.75 0.00 |
| V₂ S₃           | 59.95 46.84 30.00 21.45 14.83 8.83 0.00 0.00 |
| V₂ S₄           | 17.52 6.70 0.00 0.00 0.00 0.00 0.00 0.00 |
| V₃ S₁           | 84.16 70.18 56.53 43.28 35.45 26.80 17.08 11.01 |
| V₃ S₂           | 77.45 61.23 49.63 36.18 25.50 16.36 8.07 2.03 |
| V₃ S₃           | 63.75 52.76 44.86 32.49 24.08 14.50 3.82 0.00 |
| V₃ S₄           | 25.93 9.57 1.57 0.00 0.00 0.00 0.00 0.00 |
| V₄ S₁           | 82.37 69.05 53.99 40.91 29.02 22.64 16.92 8.40 |
| V₄ S₂           | 75.35 59.44 46.12 35.16 21.41 11.29 6.09 0.00 |
| V₄ S₃           | 63.83 48.77 35.67 26.58 19.55 11.57 3.51 0.00 |
| V₄ S₄           | 21.43 8.29 0.00 0.00 0.00 0.00 0.00 0.00 |
| S.Em. ±         | 0.51 0.53 0.61 0.53 0.53 0.36 0.27 0.10 |
| C.D. at 5%       | 1.42 1.49 1.72 1.50 1.50 1.00 0.76 0.27 |
| C.V. %           | 2.50 3.37 5.18 6.22 8.63 8.59 12.71 11.46 |
Combined interaction effect of variety, media and storage temperature on mango pollen viability

Due to combined interaction effect of variety, media and storage temperature, the wide variation was also observed significant at all days of storage (Table-5). Highest pollen viability (95.39, 86.65, 74.78, 61.64 & 49.53%) were noted in variety Mallika stored in n-hexane at -20°C (V₂M₁S₁) at 7, 14, 21, 35 and 42 days of storage, respectively. At 28 and 56 days of storage, maximum pollen viability (62.99 and 25.47%) was noted in Kesar stored in n-hexane at -20°C (V₃M₄S₄), respectively. At 49 days of storage, maximum pollen viability (35.47%) was noted in variety Dudhpendo stored in n-hexane at -20°C (V₄M₄S₄). Lowest pollen viability was noted in treatment combination V₄M₃S₄ at 7 days, V₄M₄S₄ at 14 days, V₃M₄S₄ at 21 days and V₁M₃S₂ at 28 days of storage, respectively. Likewise, lowest pollen viability was found in V₂M₄S₂ at 35, 42 and 56 days of storage, respectively. However, it was noted zero with treatment combination V₂M₃S₄ and V₄M₃S₄ from 14 days of storage. No pollen viability was observed in all varieties with no media and any of storage temperature after 28 days of storage. Variation in pollen viability was observed due to different genetic constitutions of varieties, different polar and non-polar nature of storage media and different metabolic activities of pollen at various storage temperatures.

Table 5: Combined interaction effect of variety, media and storage temperature on pollen viability

|                                       | V₁   | V₂   | V₃   | V₄   | V₅   | V₆   |
|---------------------------------------|------|------|------|------|------|------|
| At 7 days of storage                  |      |      |      |      |      |      |
| V₁                                   | M₁   | M₂   | M₃   | M₁   | M₂   | M₃   |
| S₁                                   | 92.22| 86.24| 66.07| 93.35| 81.03| 61.58|
| S₂                                   | 88.57| 80.27| 51.62| 84.31| 71.71| 52.71|
| S₃                                   | 86.79| 80.39| 40.55| 75.85| 68.81| 35.60|
| S₄                                   | 41.92| 28.40| 19.04| 26.09| 16.11| 10.37|
| Combined interaction effect of variety, media and storage temperature on pollen viability |      |      |      |      |      |      |
| At 14 days of storage                 |      |      |      |      |      |      |
| V₁                                   | M₁   | M₂   | M₃   | M₁   | M₂   | M₃   |
| S₁                                   | 85.87| 74.06| 50.16| 84.67| 71.56| 45.75|
| S₂                                   | 77.88| 71.33| 38.59| 72.68| 63.00| 36.57|
| S₃                                   | 76.16| 61.42| 30.40| 67.33| 51.58| 21.60|
| S₄                                   | 21.65| 13.54| 3.20 | 10.48| 14.39| 0.00 |
| At 21 days of storage                 |      |      |      |      |      |      |
| V₁                                   | M₁   | M₂   | M₃   | M₁   | M₂   | M₃   |
| S₁                                   | 73.35| 69.33| 36.32| 70.11| 63.53| 24.25|
| S₂                                   | 66.72| 55.86| 19.90| 59.89| 51.53| 14.33|
| S₃                                   | 62.33| 53.12| 11.59| 50.73| 39.29| 0.00 |
| S₄                                   | 8.77 | 5.40 | 0.00 | 0.00 | 0.00 | 0.00 |
| At 28 days of storage                 |      |      |      |      |      |      |
| V₁                                   | M₁   | M₂   | M₃   | M₁   | M₂   | M₃   |
| S₁                                   | 62.99| 53.42| 21.27| 56.18| 45.84| 11.54|
| S₂                                   | 52.42| 44.59| 8.72 | 43.30| 34.18| 0.00 |
| S₃                                   | 51.31| 39.50| 0.00 | 37.35| 27.01| 0.00 |
| S₄                                   | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| At 35 days of storage                 |      |      |      |      |      |      |
| V₁                                   | M₁   | M₂   | M₃   | M₁   | M₂   | M₃   |
| S₁                                   | 58.90| 44.57| 0.00 | 43.14| 34.63| 0.00 |
| S₂                                   | 42.77| 31.56| 0.00 | 32.82| 27.25| 0.00 |
| S₃                                   | 38.33| 26.34| 0.00 | 26.74| 17.76| 0.00 |
| S₄                                   | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| At 42 days of storage                 |      |      |      |      |      |      |
| V₁                                   | M₁   | M₂   | M₃   | M₁   | M₂   | M₃   |
| S₁                                   | 47.74| 31.22| 0.00 | 31.01| 22.23| 0.00 |
| S₂                                   | 26.77| 21.03| 0.00 | 22.04| 15.37| 0.00 |
| S₃                                   | 27.63| 16.69| 0.00 | 16.21| 10.29| 0.00 |
| S₄                                   | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| At 49 days of storage                 |      |      |      |      |      |      |
| V₁                                   | M₁   | M₂   | M₃   | M₁   | M₂   | M₃   |
| S₁                                   | 32.71| 19.86| 0.00 | 18.63| 11.74| 0.00 |
| S₂                                   | 20.53| 9.16 | 0.00 | 10.39| 6.87 | 0.00 |
| S₃                                   | 10.66| 0.00 | 0.00 | 11.45| 0.00 | 0.00 |
| S₄                                   | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
| At 56 days of storage                 |      |      |      |      |      |      |
| V₁                                   | M₁   | M₂   | M₃   | M₁   | M₂   | M₃   |
| S₁                                   | 32.71| 19.86| 0.00 | 18.63| 11.74| 0.00 |
| S₂                                   | 20.53| 9.16 | 0.00 | 10.39| 6.87 | 0.00 |
| S₃                                   | 10.66| 0.00 | 0.00 | 11.45| 0.00 | 0.00 |
| S₄                                   | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 | 0.00 |
Conclusion
On the basis of results obtained from the present investigation, it can be concluded that mango pollen grains could be stored successfully in different storage media at low temperature up to 56 days. Among four varieties tested, Kesar recorded as good in terms of pollen storage ability. In storage media, n-hexane was found best in terms of retaining more viability in all four varieties but on the other hand success rate was found minimum in case of no media. Storage at -20°C gave better results in terms of pollen viability under study. Therefore, the best suitable temperature for long duration of pollen storage pollen storage would be -20°C. At room temperature without media, pollen grains could not be stored more than a week due to rapid decrease in their viability. Due to that, pollen grains stored without media and at room temperature cannot be adopted. Pollen grains of Kesar stored in n-hexane at -20°C were found best during whole research experiment.

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