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**Studies on pollen viability in pomegranate (*Punica  
granatum L.*)**

**Ramandeep Kumar and Somandeep Kaur**

**Abstract**

The present investigations entitled “Studies on pollen viability in pomegranate (*Punica granatum L.*)” was conducted at pomegranate blocks, Department of Fruit Science, Dr. Y S Parmar University of Horticulture and Forestry, Nauni, Solan house during in lieu of conducting hybridization studies. Six soft seeded cultivars namely Ganesh, G-137, Dholka, Nabha, Jodhpur Red, Mridula and three hard seeded cultivars namely Kandhari Hansi, China Seedling and Bush Large were used for pollen studies. Pollen viability assessed by Acetocarmine solution (2%), Tetrazolium solution (1%) and Erythrosine B (0.1%). The pollen viability in Acetocarmine ranged from 90.79 to 97.86 per cent whereas in Tetrazolium and Erythrosine B, pollen viability varied from 88.94 to 97.13 per cent and 80.76 to 94.65 per cent respectively.

**Keywords:** Viability, pomegranate, Acetocarmine solution, tetrazolium solution and erythrosine B

**1. Introduction**

Pomegranate (*Punica granatum L.*) belongs to family Punicaceae, is a favourite table fruit of the tropical and subtropical regions. It is very much liked for its cool refreshing juice and valued for its medicinal properties. It is an ancient fruit which has been originated in Persia, Afghanistan and Baluchistan (De Candolle, 1967) [2]. The exact date of introduction in India is not known but, it was established and became naturalized very early in Western India (Prayag, 1940) [5]. Various cultivars have been recommended for commercial cultivation in India *i.e.* Ganesh and Muscat in Maharashtra, Dholka in Gujrat, Jalor Seedless, Jodhpuri Red, Jodhpuri White and Seedless Bedana in Rajasthan, Bassien Seedless and Madhurigi in Karnataka, Chawla, Nabha, Country Large Red in Haryana (Hisar) and Srinagar Special in valley areas of Garhwall hills). The total area under pomegranate cultivation in Himachal Pradesh is 1085 hectare with the production of 475 MT during 2009 (Anonymous, 2009) [1]. The total area under cultivation of pomegranate in India is 125 thousand hectares and production is around 820.30 thousand MT during 2010 (NHB, 2010) [3]. Pomegranate plant can withstand frost (temperature up to -10 °C) and can grow up to an altitude of 1600 meters above mean sea level (Rana and Dwivedi, 1997) [6]. Thus, pomegranate which has high market demand, longer shelf-life, versatile adaptability, hardy nature, low maintenance cost, steady and high yield can provide a better alternative for stone fruits (Patil *et al.*, 2004) [4]. Flowering time, pollen characters and the morphological features of flowers are useful in the classification of different cultivars. The acceptability of a cultivar by the consumers depends largely on its fruit quality. Therefore, with a view to infusing blood red aril colour and soft seededness in some of the hard seeded commercial cultivars like Kandhari Hansi, the present investigations were carried out involving Ganesh, G-137, Dholka, Nabha, Jodhpur Red, Mridula, Kandhari Hansi, China Seedling and Bush Large with the following objectives:

To determine pollen viability between different cultivars.

**2. Materials and Methods**

The present investigations were carried out in Dr. Y S Parmar University of Horticulture and Forestry, Nauni, Solan (H P). The cultivars undertaken for studies were Ganesh, G-137,

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Dholka, Nabha, Jodhpur Red, Mridula, Kandhari Hansi, China Seedling, Bush Large. The methodology used in present work is represented as under.

## 2.1 Pollen viability

### 2.1.1 Acetocarmine test (2%)

Pollen grains were stained in 2 per cent Acetocarmine solution. Acetocarmine solution was freshly prepared by taking 45 per cent of glacial acetic acid and 2 grams of carmine powder and was dissolved to make final volume 100 ml by adding distilled water. The mixture was boiled for five minutes and then filtered with Whatman No.1 filter paper. The pollen grains were dusted on a glass slide and one to two drops of acetocarmine solution were put on these grains, covered with the cover slip and was left for 4-5 minutes for proper staining. Slides were examined under microscope. Deeply stained and normal looking pollen grains were considered to be viable, whereas shriveled; lightly stained or colorless pollen grains were counted as non-viable. Three different fields were observed and viable, non-viable pollen grains were counted in each field under the simple binocular microscope.

### 2.2 Tetrazolium test (1%)

Tetrazolium solution was prepared by dissolving one gram of Tetrazolium salt in distilled water to make final volume 100 ml. Pollen grains were dusted on glass slides and one to two drops of Tetrazolium solution (1%) were put on them. Cover slip was placed over it and slides were kept as such for 20-25 minutes. Observations were recorded in three different fields under microscope.

### 2.3 Erythrosine B test (0.1%)

Erythrosine B solution (0.1%) was prepared by dissolving 0.1gram of Erythrosine B in distilled water to make a final solution of 100 ml. The unstained pollen grains were considered viable in this method and stained pollen grains were considered as non-viable.

## 2.4 Statistical Analysis

Replications : 3

CRD : Pollen viability

The data was analyzed as per the procedures given by Panse and Sukhatme (1985) [7].

## 3. Results and Discussion

The results obtained from present investigations are

### 3.1 Acetocarmine test (2%)

Pollen viability recorded in different pomegranate cultivars are presented in (Table 4.1). The data presented in table

reveals that pollen viability of various cultivars differed significantly, when pollen grains were stained with Acetocarmine solution (2%). Maximum pollen viability (97.86%) was observed in G-137, which was found at par with Bush Large (97.79%), Ganesh (97.36%). Pollen viability in China Seedling was recorded 96.68 per cent followed by Kandhari Hansi (96.04%), Nabha (95.77%), Jodhpur Red (94.63%) and Mridula (90.79%). Minimum pollen viability of 85.27 per cent was observed in Dholka. The colour of pollen grains was observed as light to deep pink after staining with Acetocarmine solution (2%).

### 3.2 Tetrazolium test (1%)

Significant variations were observed (Table 4.1) in pollen viability when Tetrazolium salt (1%) solution was used. Maximum pollen viability (97.13%) was recorded in Bush Large followed by Jodhpur Red having 96 per cent pollen viability. Minimum pollen viability (88.49 %) was recorded in Mridula, which was significantly lower than pollen viability of other cultivars.

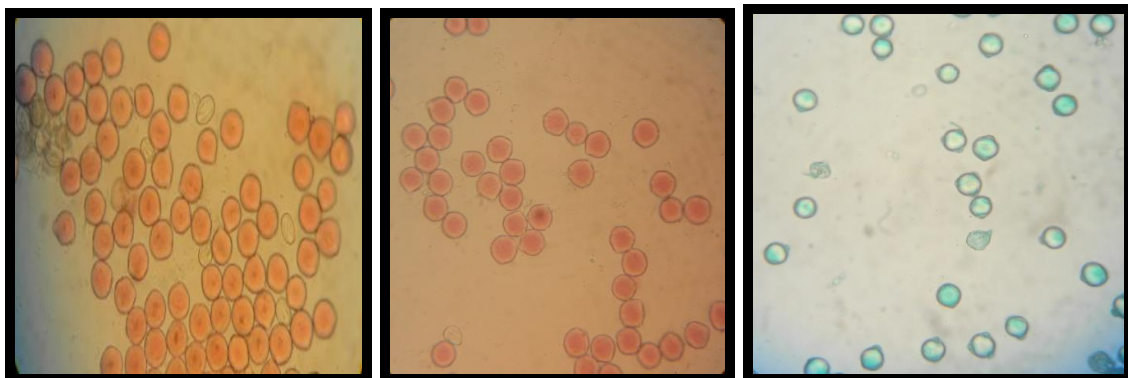
Pollen viability in G-137 and Ganesh was 95.41 and 94.10 per cent respectively. Dholka (93.72%) was found at par with China Seedling having 93.45 per cent pollen viability. Pollen viability recorded in Nabha (92.28%) was observed at par with Kandhari Hansi (91.90%).

### 3.3 Erythrosine B test (0.1%)

Pollen viability was found (Table 4.1) highest in Kandhari Hansi (94.65%) followed by Mridula (92.20%) and least in China Seedling (78.92%), when tested with 0.1 per cent Erythrosine B. It was found that Ganesh (85.67%) was statistically at par with G-137 (85.15%) and Nabha (84.39%) and Dholka (83.81%). Pollen viability in Bush Large and Jodhpur Red was recorded 88.75 and 80.76, per cent respectively.

**Table 4.1:** Pollen viability of pomegranate cultivars with different staining solutions

Cultivars	Acetocarmine (2%)	Tetrazolium (1%)	Erythrosine B (0.1%)
Kandhari Hansi	96.04 (9.79)*	91.90 (9.58)	94.65 (9.73)
China Seedling	96.68 (9.83)	93.45 (9.67)	78.92 (8.89)
Bush Large	97.79 (9.88)	97.13 (9.86)	88.75 (9.42)
Dholka	85.27 (9.23)	93.72 (9.68)	83.81 (9.15)
Jodhpur Red	94.63 (9.73)	96.00 (9.80)	80.76 (8.99)
Ganesh	97.36 (9.87)	94.10 (9.70)	85.67 (9.26)
G-137	97.86 (9.89)	95.41 (9.77)	85.15 (9.22)
Nabha	95.77 (9.79)	92.28 (9.60)	84.39 (9.19)
Mridula	90.79 (9.53)	88.49 (9.40)	92.20 (9.60)
Mean	94.69 (9.73)	93.61(9.67)	86.03 (9.27)
CD <sub>0.05</sub>	0.12	0.25	0.39



**Fig 1:** Slides and figures of pollen viability in acetocarmine (2%), Erythrosin B(1%) and tetrazolium salt solution(0.1%)

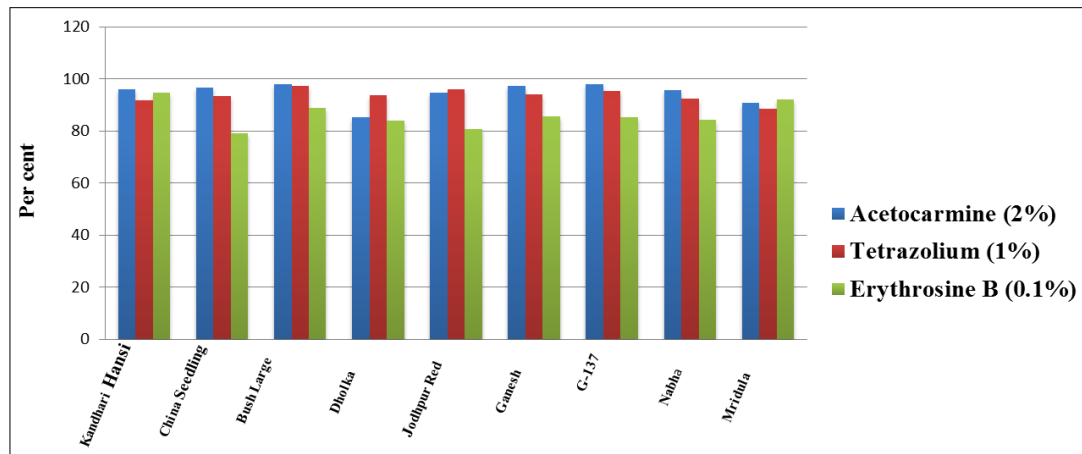


Fig 2: Cultivars

#### 4. Conclusion

The investigations concluded that Pollen viability was found highest in G-137 by Acetocarmine test and in Bush Large by Tetrazolium test. In Erythrosine B test Kandhari Hansi has highest pollen viability.

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