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# Studies on pollen quality and quantity, stigma receptivity, pollination and fruit set in raspberry (*Rubus paniculatus* S.) wild species of Garhwal Himalaya, Uttarakhand, India

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#### Abstract

Raspberry wild species (*Rubus paniculatus* S.) of Garhwal Himalaya was selected to determine the pollen quality and quantity, pollination and fruit set under the hilly regions of Garhwal Himalaya. The raspberry produced 1,59,012 number of pollens per flower and exhibited maximum pollen grain size (length x width) of  $35.87 \times 17.73 \mu$  in dry conditions and  $35.44 \mu \times 16.52 \mu$  in glycerine. The pollen viability was 73.33% and its longevity was extended up to 7th day. Pollens showed delayed germination (24 hours), the treatment T<sub>16</sub> (25% Sucrose + 5ppm Gibberellic acid) showed maximum pollen germination 9.66%, 37.82% and 52.34% and pollen tube length of  $88.05 \mu$ ,  $99.18 \mu$  and  $135.22 \mu$ , at 24, 48 and 78 hours after planting. Stigma receptivity was highest on the day of anthesis, pollination studies indicated that maximum fruit set was observed on natural pollination (cross pollination) than any other method of pollination. Interspecific pollination of *Rubus paniculatus* S. with *Rubus niveus* T. and *Rubus macilentus* C. revealed that *Rubus paniculatus* S. is good male parent with better fruit set, when it was used as female parent, did not set any fruits.

Keywords: Pollen viability, germination, stigma receptivity, fruit set, pollination

#### Introduction

The genus *Rubus* belongs to family Rosaceae, with a large number of highly variable and heterogeneous species of raspberries and blackberries, which occur in all parts of the world except dessert regions. All raspberries are included in the subgenus *Idaeobatus*. Even though India has wide genotypic diversity of raspberry with nutritional and medicinal properties, commercial orchards are seen due to negligible area and due to lack of improved varieties. In this context present study gives the basic information to the breeders for crop improvement. It is well known that the fruit set and yield are strongly depends upon pollen quantity and qualities, stigma receptivity, mode s of pollination, Effective pollination period (EEP) and fruit set and retention play a critical role. There is little literature available about flowering, pollen studies, pollination and fruit set. In addition to that it is expected to give information on protecting *Rubus paniculatus* S. from endangered population.

#### **Materials and Methods**

The present experiment was carried out at VCSG. Uttarakhand University of Horticulture and Forestry, Bharsar, Pauri Garhwal, Uttarakhand during 2016-2017.

#### **Pollen Studies**

The flower buds at balloon stage were collected on the previous evening of anthesis day from Rubus paniculatus S. plants selected for studies during peak period of flowering and the flower buds were placed in vase with water. To determine quantity of pollens per anther and flower, the pollens were placed in 1 ml of glycerine 1%, in a small vial, anthers were crusherd and the pollen grains were released in to the solution. From this mixture, five (10  $\mu$ l) droplets were taken out in the slides and examined under the microscope, to determine the pollens per anther and pollen grains per flower was estimated by multiplying the number of pollen grains per anther with total number of anthers per flower.Studies on pollen morphology under different medium (Table-2) by taking the average size (length and breadth) of 50 pollen grains measured under compound microscope in each medium viz., dry condition, water,

acetocarmine and glycerine with the help of ocular micrometer indexed against stage micrometer to determine the final value. Fresh pollen grains were subjected to acetocarmine test to study pollen viability and longivity (Table-3). The stained pollens were considered as viable pollens, the same tests were continued with the flowers collected and stored at room conditions (25<sup>o</sup> C) on alternative days to estimate longivity of pollens. The pollen viability was calculated by dividing number of stained pollen grains by total number of pollen grains observed and multiplying the resulted value with 100 and pollen germination and pollen growth under different artificial medias at different time intervals (Table-4 and 5) as suggested by Shivanna and Rangaswamy (1992) <sup>[11]</sup> was carried out. Pollen germination percentage was calculated by dividing Number of germinated pollen grains with Total number of pollen grains observed and multiplying the product value with 100 and pollen tube length was measured using ocular micrometer indexed against stage micrometer to arrive the final value. The studies were The laboratory experiment were laid out in the Randomized Block Design with three replications for pollen load, viability, longevity and three replications and 16 treatments for Pollen germination and pollen tube growth, while six replications and four treatments were used for pollen morphology studies. The data collected on various parameters were analyzed using the software OPSTAT to find out the significance of variation resulting from the experimental treatments. The mean values were subjected to analysis of variance as per Statistical Procedure for Agricultural Research (Gomez and Gomez. 1983<sup>[7]</sup>) for Randomized Block Design.

# Stigma Receptivity

The stigma receptivity was studied by two methods, viz., visual observations and fruit-set method. The stigmas of different age groups were observed daily morning from three days prior up to three days after anthesis, with the help of hand lens. The stigmas looking shiny, sticky, fresh and attractive were considered as receptive while dull, faded, non sticky and brownish stigmas were considered as non-receptive and in fruit set method, 10 emasculated flower buds were hand self pollinated at different ages as selected in visual method. The pollinated buds were covered with paper bags and tagged as usual. The fruit set may observe 21 days after pollination which may be indicated by initiation of swelling of ovaries, calvx cup covering back the ovaries, weathering of stamens and the fruit set percentage was calculated by dividing Number of fruits set with Total number of pollinated flowers multiplying the value with 100.

# **Pollination Studies**

The different methods were employed to study the mode of pollination in raspberry wild species. Allogamy was studies by selecting Mature flower buds, ready to open next day were emasculated and pollination was done on next day (at anthesis) with the pollens collected from the freshly dehisced anthers of another plant or same species, these pollinated flowers were bagged again and allowed to remain on the plants for fruit set. Interspecific crosses between Rubus macilentus C. X Rubus paniculatus S., Rubus niveus T. X Rubus paniculatus S., Rubus paniculatus S. X Rubus macilentus C. and Rubus paniculatus S.X Rubus niveus T. were carried out. To study Autogamy the shoots bearing flower buds were tagged and bagged with all necessary care, a day before anthesis and left for self-pollination and fruit-set inside the bags. Geitonogamy was studied in the same way as that of allogamy except the pollens selected for pollination were from same plant. Natural or open pollination was studied by selecting Perfect flower buds on ten plants of each raspberry wild species were tagged before anthesis and allowed to remain on the trees for recording various observations with respect to fruit-set. Self incompatibility was obtained by dividing the average fruit set after self pollination by the average fruit set after cross pollination (Lloyd and Schoen, 1992) <sup>[8]</sup>. The value of one indicates complete self incompatibility. Anthers and stigma of buds were clipped a day prior to anthesis and bagged. If there is fruit set then it was considered as an apomictic fruit under apomixes studies.

# Fruit Set

The time and percentage of fruit set in the raspberry wild species was carried out by selecting five branches on the periphery of a plant during flowering season. The flowers on each branch were counted. The observation on fruit set was started after one week of natural pollination. The date on which percentage of fruit set recorded is 75per cent or above was considered as the time of fruit set. After three weeks of open pollination, fruit set in each branch was recorded and the fruit set per 100 flowers was calculated in order to get percentage of fruit set in all the raspberry wild species under study and Percentage of fruit set was calculated by dividing Number of fruits set with Total number of flower counted and the resulting value was multiplied by 100.

### **Results and Discussion Pollen Studies**

**Number of pollens per anther:** Pollen quantity is one of the essential factors for effective pollination and fruit set. 1100 pollens per anther and 1,59,012 pollens per flower was found in a hermaphrodite flower of *Rubus paniculatus* S. indicating good amount of pollen load for pollination and the values are given in the Table-1 the similar studies were made by Xie *et al.* (2003) <sup>[12]</sup>, he observed 8428 and 1197 pollen grains per anther in apple and japanese plum respectively.

**Table 1:** Number of pollens per anther and per flower.

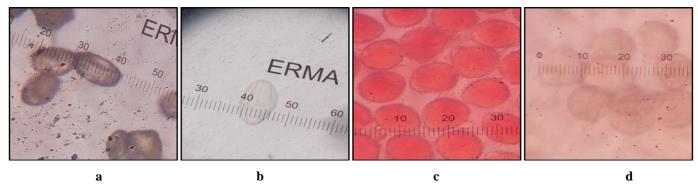
Observation	Rubus paniculatus S.
Number of pollens per anther	1260±114.93
Number of pollens per flower	1,59,012

Morphology of pollen grains in different conditions: Pollen grains were pale yellow but in mass pollen grains looked golden yellow in color. The shape of pollen grains in glycol and dry conditions were oblong, and ellipitic, while in acetocarmine and water pollen grains appeared to be round in shape. The values are given in Table-2, on an average the maximum pollen grains size (length x width) of 35.87 x 17.73  $\mu$  was noticed in dry conditions, followed by glycerine (35.44  $\mu \ge 16.52 \mu$  (plate-1). The minimum size of pollen grains  $(27.89 \ \mu \ x \ 26.92 \ \mu)$  was recorded under the acetocarmine conditions, followed by water (29.69  $\mu$  x 28.54  $\mu$ ). The present results are inline with the findings of Pawar et al. (2017) <sup>[10]</sup> the pollen grains of raspberry (*Rubus ellipticus*) exhibited the average size (length X width) of 177.73  $\mu$  X 124.43  $\mu$  in glycerol condition, followed by 173.29  $\mu$ X173.29  $\mu$  in water suspension and the shape of pollen gains in glycol and dry conditions were oblong, elliptic and in acetocarmine and water pollen grains appeared to be round in shape.

Treatment	<i>Rubus paniculatus</i> S. pollen ± SE(m)			
Treatment	Length (µ)	Width (µ)		
T1 - Control (Water)	29.69±0.48	29.98±0.52		
T2- Glycerine	35.44±0.27	16.52±0.39		
T3 - Acetocarmine	27.89±0.49	26.92±0.47		
T4 – Dry condition	35.87±0.43	17.73±0.39		
SE(d)	0.60	0.63		
C.D.	1.27	1.33		
C.V.	3.25	4.82		

Table 2: Morphology of pollen grains in different conditions

 $\mu$  - micron (unit of pollen tube length)



**Plate 1:** Morphology of pollens under different conditions (a. Under dry condition b. Glycerine c. Acetocarmine d.Water)

**Pollen viability and longevity:** The viability of pollen grains of *Rubus paniculatus* S. was 73.33% and the pollen longevity was extended up to 7<sup>th</sup> day with 16.66% and became nil on 9<sup>th</sup> day of storage of pollen grains under room conditions(Table-3). Otterbacher *et al.* (1983) <sup>[9]</sup> showed that high temperature resulted in rapid loss of viability of raspberry pollen. The

germinability of pollen grains stored under room conditions indicated that after three days of storage, the pollen grains were quite normal and showed 46.66% germination. But after seven days of storage, the germinability was decreased rapidly and after nine days of storage, the germination percentage totally declined.

Table 3: Viability and longevity of pollens

Species	Longevity of pollens at different days intervals				
Species	1 <sup>st</sup> day	3 <sup>rd</sup> day	5 <sup>th</sup> day	7 <sup>th</sup> day	9 <sup>th</sup> day
Rubus paniculatus S.	73.33%	70.00%	45.33%	16.66%	0.00%

**Pollen germination :** The results (Table- 4 and 5) indicated that *Rubus paniculatus* S. showed delayed germination (24 hours), hence the observations were made at 24, 48 and 72 hours after planting of pollen grains in artificial media of sucrose, boric acid and gibberellic acid at different concentration combinations and water served as control. The treatment T<sub>16</sub> (25% Sucrose + 5ppm Gibberellic acid) showed maximum pollen germination 9.66%, 37.82% and 52.34% and pollen tube length of 88.05  $\mu$ , 99.18  $\mu$  and 135.22  $\mu$ , at 24, 48 and 78 hours after planting at room conditions (25<sup>o</sup> C). It was observed that all the treatments at different intervals of time (24, 48 and 72 hours) were significant for pollen germination and pollen tube length when compared to control (water) The pollen germination in shown in Plate-2.

After 24 hours of pollen planting, the maximum pollen germination percentage (9.66%) was recorded with  $T_{16}$  (25% Sucrose + 5ppm Gibberellic acid). The minimum pollen germination percentage (5.35%) recorded with  $T_2$  and no germination was seen in  $T_1$  control (Water). The maximum pollen tube length (88.05  $\mu$ ) was recorded with  $T_{16}$  (25% Sucrose + 5ppm Gibberellic acid) which was statistically at par with treatment  $T_6$ ,  $T_9$ ,  $T_{10}$ ,  $T_{11}$  and  $T_{15}$  recording 81.57  $\mu$ , 75.43  $\mu$ , 80.35  $\mu$ , 77.31  $\mu$  and 76.22  $\mu$  respectively. Minimum pollen tube length (0  $\mu$ ) was recorded  $T_1$ - Water (Control). Observations on taken after 48 hours of pollen planting resulted in maximum pollen germination percentage (37.82%)

and pollen tube length (99.18  $\mu$ ) was recorded with T<sub>16</sub> (25% Sucrose + 5ppm Gibberellic acid). The maximum pollen tube length was recorded with T<sub>16</sub> (25% Sucrose + 5ppm Gibberellic acid) which was statistically at par with treatment T<sub>11</sub>, T<sub>14</sub>, T<sub>15</sub>, T<sub>6</sub> and T<sub>10</sub> recording 89.37  $\mu$ , 89.26  $\mu$ , 90.05  $\mu$ , 88.21  $\mu$  and 88.61  $\mu$  respectively. The minimum pollen germination percentage (3.05%) and tube length (0  $\mu$ ) was recorded with T<sub>1</sub> (Water).

The data on pollen germination and pollen tube length in raspberry (*Rubus paniculatus* S.) was observed after 72 hours of pollen incubation and the results are presented in Table 4 and 5. The data revealed that the maximum pollen germination percentage (52.34%) was recorded with  $T_{16}$  (25% Sucrose + 5ppm Gibberellic acid). The minimum pollen germination percentage (9.68%) was recorded with  $T_1$  (Water). The maximum pollen tube length (135.22 $\mu$ ) was recorded with  $T_{16}$  (25% Sucrose + 5ppm Gibberellic acid) which was statistically at par with treatment  $T_{14}$  (124.38  $\mu$ ). Minimum pollen tube length (14.18 $\mu$ ) was recorded  $T_1$  (Water).

Pawar *et al.*(2017) <sup>[10]</sup> reported 25% sucrose + 0.4% boric acid media was recorded maximum pollen germination of 36.66% and pollen tube length of 284.37  $\mu$  after 48 hours of planting. The minimum pollen germination 4.33% and pollen tube length 13.33  $\mu$  was recoded in water after 48 hours of planting. Abdel (1999) <sup>[1]</sup> reported pollens planted in 15 per

cent sucrose solution resulted in 73.30 to 86.10 per cent pollen germination in three apple cultivars, but best pollen

tube growth (26.66  $\mu$  to 284.37 $\mu$ .) was observed with 25% sucrose + 0.4% boric acid solution.

Table 4: Pollen	germination	percentage of I	Rubus	paniculatus S.	after	different	time of incubation
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Treatments	Pollen germination (%)±SE(m) (24hours)	Pollen germination (%)±SE(m) (48hours)	Pollen germination (%)±SE(m) (72hours)
T1- Water(Control)	0.00±0.00	$3.05 \pm 0.08$	9.68±0.16
T2- 0.1% Boric acid + 5% Sucrose	5.35±0.11	22.63±0.11	31.73±0.08
T3 - 0.2% Boric acid + 10% Sucrose	5.62±0.16	23.75±0.12	38.81±0.09
T4- 0.3% Boric acid + 15% Sucrose	7.80±0.14	28.14±0.13	40.56±0.05
T5- 0.4% Boric acid + 20% Sucrose	9.52±0.17	31.57±0.13	45.8±0.06
T6 - 0.5% Boric acid + 25% Sucrose	8.25±0.13	31.46±0.11	30.67±0.09
T7 - 0.1% Boric acid + 1ppm Gibberellic acid	6.59±0.15	22.61±0.09	32.33±0.08
T8 - 0.2% Boric acid + 2ppm Gibberellic acid	7.80±0.09	24.57±0.14	35.85±0.06
T9 - 0.3% Boric acid + 3ppm Gibberellic acid	8.42±0.10	25.69±0.13	38.75±0.07
T10 - 0.4% Boric acid + 4ppm Gibberellic acid	8.89±0.08	27.56±0.10	41.84±0.05
T11 - 0.5% Boric acid + 5ppm Gibberellic acid	8.06±0.06	28.29±0.13	42.78±0.08
T12 - 5% Sucrose + 1ppm Gibberellic acid	6.88±0.08	27.75±0.11	39.95±0.04
T13 - 10% Sucrose + 2ppm Gibberellic acid	6.94±0.06	30.68±0.07	43.78±0.06
T14 - 15% Sucrose + 3ppm Gibberellic acid	7.67±0.09	34.83±0.08	45.58±0.07
T 15 - 20% Sucrose + 4ppm Gibberellic acid	9.07±0.08	36.71±0.07	49.09±0.08
T16 - 25% Sucrose + 5ppm Gibberellic acid	9.66±0.07	37.82±0.09	52.34±0.07
SE(d)	0.15	0.15	0.11
CD <sub>0.05</sub>	0.31	0.32	0.23
C.V.	2.54	0.69	0.36

Table 5: Pollen tube length of Rubus paniculatus S. after different time of incubation

Treatments	Pollen tube length (µ)±SE(m) (24hours)	Pollen tube length (µ)±SE(m) (48hours)	Pollen tube length (µ)±SE(m) (72 hours)
T1- Water(Control)	0.00±0.00	$0.00 \pm 0.00$	$14.18 \pm 0.77$
T2- 0.1% Boric acid + 5% Sucrose	35.51±1.7	40.48±1.57	37.51±4.60
T3 - 0.2% Boric acid + 10% Sucrose	52.70±3.06	60.65±6.63	57.09±5.37
T4- 0.3% Boric acid + 15% Sucrose	62.43±3.64	61.93±6.52	57.07±6.33
T5-0.4% Boric acid + 20% Sucrose	69.88±4.97	73.73±5.04	77.47±3.68
T6 - 0.5% Boric acid + 25% Sucrose	81.57±2.65	88.22±5.31	83.72±5.04
T7 - 0.1% Boric acid + 1ppm Gibberellic acid	62.13±8.40	63.51±4.70	69.72±7
T8 - 0.2% Boric acid + 2ppm Gibberellic acid	66.71±5.90	71.32±5.75	80.04±7.34
T9 - 0.3% Boric acid + 3ppm Gibberellic acid	75.43±5.71	78.07±3.38	82.95±3.39
T10 - 0.4% Boric acid + 4ppm Gibberellic acid	80.35±2.24	88.61±3.05	93.36±4.74
T11 - 0.5% Boric acid + 5ppm Gibberellic acid	77.32±4.76	89.37±4.68	94.15±7.29
T12 - 5% Sucrose + 1ppm Gibberellic acid	63.54±6.65	74.09±4.31	92.63±9.6
T13 - 10% Sucrose + 2ppm Gibberellic acid	69.72±7.00	80.58±6.56	102.19±6.27
T14 - 15% Sucrose + 3ppm Gibberellic acid	74.32±5.65	89.26±6.33	124.38±6.63
T 15 - 20% Sucrose + 4ppm Gibberellic acid	76.22±4.30	90.05±1.88	109.79±4.97
T16 - 25% Sucrose + 5ppm Gibberellic acid	88.05±4.34	99.18±4.27	135.22±5.37
SE(d)	6.94	6.75	8.28
CD0.05	14.21	13.81	16.95
C.V.	13.13	11.51	12.37

μ - micron (unit of pollen tube length)

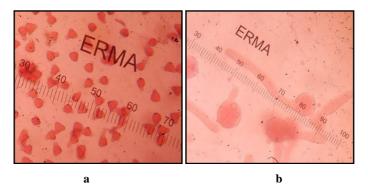


Plate 2: Pollen germination:

**a**. Initiation of pollen grain germination **b**. Germinated pollen grain with pollen tube

# Stigma Receptivity

Stigma receptivity by visual method: In order to study stigma receptivity of raspberry wild species by visual observation, the stigmas of different age groups from three days prior up to 3 days after anthesis was recorded with the help of a hand lens. Stigma which were plump and shiny with stigmatic secretion were considered to be receptive while those having dull brownish appearance and lacking in secretions were considered to be non receptive. Three days before anthesis, the stigma was white with pinkish margins, short, prominent and there was no presence of any sugary secretion on the surface, the style was more curved and fused at the centre of flower. Two days before anthesis, the stigmatic surface was little bigger than the previous day and the surface was more whitish, prominent than before. There was slight appearance of some sugary secretion on the surface of the stigma and the styles were less curvy than before but still fused. One day before anthesis the stigmatic surface was at its maximum length and start showing little amount of sugary secretion. The stigmatic surface starts to turn light creamish colour and styles became slightly free. The characteristics of receptive stigma, pollinated pistils are given in Plate-3.

On the day of anthesis, the stigmatic surface completely turned to creamish colour. It showed the highest amount of sugary secretion and fresh appearance. The pollination takes place at this stage leads to the highest amount of fruit set, the styles became completely straight and was showing maximum receptivity. One day after anthesis, flower showed the little shrunken stigmatic surface. The colour of the stigma was still creamish in colour and there was a depleted amount of sugary secretion on the surface of the stigma, again the styles became slightly curvy. Two day after anthesis, stigma shows the more shrunken stigmatic surface. The colour of the stigma was still creamish in colour and there was a depleted amount of sugary secretion on the surface of the stigma and styles were curvier than before. Three day after anthesis, stigma shows the more shrunken stigmatic surface. The colour of the stigma is started to turn from creamish to brown colour and there is a depleted amount of sugary secretion on the surface of the stigma and it starts to dry and curved styles.

**Fruit set method:** The stigma receptivity assessed through fruit set method by selecting the different flower buds of different age groups were emasculated and pollinated with fresh pollen. The flower is highly cross pollinated in nature. Fruit set was not observed on self pollination, therefore stigma receptivity by fruit set method of *Rubus paniculatus* S. was not successful. Bekey and Lawrence (1985)<sup>[2]</sup> reported that the peak receptivity period of eight cultivars of red raspberry lasted from 1 to 4 days. Most cultivars set as well with one day of hand pollination at the peak of the receptivity period as with three days of consecutive pollination.

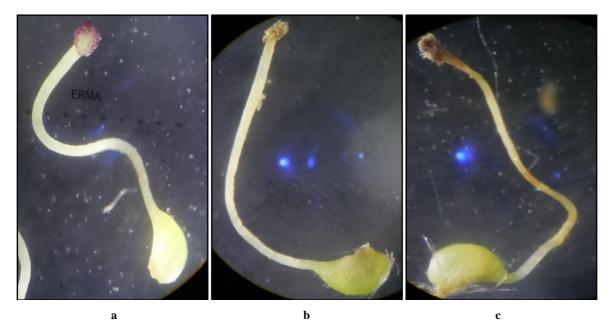


Plate 3: Stigma receptivity by visual methods a. Stigma receptivity with sugary and sticky surface b. Pollinated pistil c. Pollinated pistil with swollen ovary

#### **Pollination Studies**

In raspberry flowering the pollination mainly mediated through honey bees and ants hence, cross pollination was naturally observed. The plants were allowed for natural pollination with mediators like honey bees or ants, which reveals that the pollinators play a key role in better fruit set increments over any other methods of pollination. It was observed that maximum of 88.93% of fruit set in *Rubus paniculatus* S., where the Autogamy (Self pollination), Geitonogamy and Apomixes methods of pollination carried out under study were failed. After third day of natural pollination of *Rubus paniculatus* S. clumping of stigmas were seen upon which the pollens were germinating.

The interspecific crosses of *Rubus paniculatus* S. was carried out with wild raspberry species of *Rubus niveus* T. and *Rubus macilentus* C. of Garhwal Himalaya. The maximum fruit set was recorded on crossing between *Rubus niveus* T. X *Rubus paniculatus* S. (72.06%) and *Rubus macilentus* C. X *Rubus paniculatus* S. (56.75%). No fruit set was seen when *Rubus paniculatus* S. was used as female parent with in *Rubus macilentus* C. and *Rubus niveus* T. indicating *Rubus paniculatus* S.was compatible with *Rubus macilentus* C. and *Rubus niveus* T. only as good pollen donor (Table-6). The pollination methods failure in *Rubus paniculatus* S. may be due to self incompatibility as well as cross incompatibility with the species under study. The fruit set was seen on natural pollination it might be due to partenocarpy or pollens might be coming from fourth compatible species apart from the species under study. The actual mechanism is yet to be study. The present findings are similar to Chaudhari (1966)<sup>[6]</sup> who reported that raspberries in India have zero to five percent self-pollination, though these are naturally cross-pollinated plants. The present results are inline with the finding of Bors

and Sullivan (2005) <sup>[5]</sup> who did the crosses of *Fragaria* moschata × *Fragaria* nubicola, *Fragaria* moschata × *Fragaria* viridis, and their reciprocal combinations were done to create tetraploids for eventual introgression into octoploid cultivars of *Fragaria* × ananassa, which indicate the cross compatibility of the *Fragaria* species.

Table 6: Inter specific crosses

Interspecific crosses	Percentage of fruit set
Rubus macilentus C. X Rubus paniculatus S.	56.75
Rubus niveus T. X Rubus paniculatus S.	72.06
Rubus paniculatus S. X Rubus macilentus C.	0.00
Rubus paniculatus S. X Rubus niveus T.	0.00

# Fruit Set

# Time and percentage of fruit set

Observation on the time of fruit set revealed that *Rubus* paniculatus S. set fruits from third week of May to last week of June and it took 23 days from commencement of flowering to first fruit set. Average initial fruit set percentage of *Rubus* paniculatus S. was 68.75% of initial fruit set over an average

of 492.36 numbers of flowers per branch (Table-7) but the fruit retention was 88.71% till the harvest. Bhartiya (1980)<sup>[3]</sup> reported a time period of 7 to 12 days after full bloom for fruit set in Royal Red, Top Red and Hardeman apple cultivars. Bist (1985)<sup>[4]</sup> obtained as much as 22.40 per cent fruit retention up to maturity under cross pollination.

Table	7:	Fruit	set	percentage
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Species	Average number of flower per branch	Percentage of initial fruit set	Fruit retention
Rubus paniculatus S.	492.36	68.75%	88.71%

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