

## Studies on Pollen Viability of Pollen Parent and Stigma Receptivity of Seed Parent in Hybrid Seed Production of Tomato Under Shade House

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**ABSTRACT:** A shade house experiment was carried out to study the pollen viability of pollen parent and stigma receptivity of seed parent in hybrid seed production of tomato under shade house at Hi-Tech Horticulture unit, Saidapur farm and seed quality analysis at seed quality testing and research laboratory National Seed Project, UAS Dharwad, during 2012-13, The experiment was laid out in female and male block system separately and totally 1200 flower buds were crossed, among which 20 flower buds were crossed with respect to each treatment in each replication and different colour threads were tied to the pedicel of the crossed buds as per the treatment for easy identification. The experiment had two factors with 4 and 5 treatments respectively in three replications, factor one pollen viability of pollen parent (P) and factor two - stigma receptivity of seed parent (R). Two day-stored pollen treatment P2 exhibited significant differences for various parameters such as fruits set (67.68%), fruit weight (95.13g), number of seeds per fruit (114.83), seed weight per fruit (0.38g), seed weight per plant (7.44g) and seed recovery (0.40%). Germination percentage (92.40%), root length (9.7), shoot length(8), seedling vigour index (1648) and seedling dry weight ( $5.50 \text{ mg}^{-10}$ ) were significantly higher when used for crossing. Pollination two days after emasculation recorded significantly higher fruits set (67.68%), fruit weight (95.13g), number of seeds per fruit (114.83), seed weight per fruit (0.38g), seed weight per plant (7.44g), seed recovery (0.40%) germination (94.33%), seedling vigour index (1760) and seedling dry weight ( $5.86 \text{ mg}^{-10}$ ) followed by R<sub>3</sub>-Pollination three days after emasculation, R<sub>1</sub>-Pollination one day after emasculation, R<sub>4</sub>-Pollination four days after emasculation and R<sub>0</sub>-Pollination on the same day of emasculation.

**Key words:** Shade House, Hybrid seed production techniques, Pollen viability and Stigma receptivity

### INTRODUCTION

Tomato (*Solanum lycopersicum* L.) belongs to the family Solanaceae (also known as the nightshade family) having chromosome number ( $2n=24$ ). It is believed to have originated from Peruvian and Mexican region. It is one of the most popular self pollinated and widely grown annual vegetable crops. It is being cultivated in kitchen gardens, commercial fields and under polyhouse, shade house, net house, poly tunnel condition and soil less culture or hydroponic systems.

Shade house is an important low cost structure for the farmers, to minimize biotic and

abiotic stress on seed crops, which could be grown round the year, during off-season and also under extreme climatic conditions. The shading is effective in reducing light penetration and temperature inside the shade house, thereby creating better microclimate for production of fruits and seeds with higher yield and quality. [1].

The average seed yield of tomato hybrid seed production is influenced by several aspects. Although, several studies have been carried out in tomato to enhance its average commercial yield in open field condition, it is important to

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search for alternatives for growing healthy and productive crop in a species like tomato where hand emasculation and hand pollination is used commonly in India. The shade house structures are identified as the best option for protection against the environmental adversities that may hamper the manual processes involved in tomato hybrid seed production..

Pollen is the precious genetic material which transmits the male gametic material during sexual reproduction of all higher plants. Hybrid seed production involves an effective crossing of emasculated flower with desired pollen or sterile female parent with a selected fertile restorer. [2] reported that the pollen can be stored for three to four days under ordinary conditions and [3] reported that two days old pollen is best to achieve the highest fruits set (%) and seedling vigour index in DTH-1 tomato hybrid female parent line. Further, in hybrid seed production inadequacy of pollens on account of biotic or abiotic factors is very much felt and it leads to poor seed set. Hence the protected structures like shade house can play a vital role in producing healthy pollen. There is a need to acquire knowledge on pollen viability of the plants which are produced under shade house condition.

Optimal receptivity of the style at pollination time is of utmost importance for efficient hybrid seed production and the desirable quantity of hybrid seeds can be obtained from the female parent when there is a perfect coincidence of stigma receptiveness and pollen viability. The stigma receptivity also plays a crucial role in hybrid seed production of tomato; [4] reported that the stigma becomes receptive 16 hours before anthesis and receptivity is retained for two to three days after anthesis. [3] reported that highest fruit set percentage and seedling vigour index were observed when pollination is done three days after emasculation in Arka Alok tomato seed parent. The stigma receptivity is largely influenced by the biotic or abiotic factors which can be manipulated by use of protected structures like shade house. Therefore, stigma receptivity after the days of emasculation and subsequent pollinations, is to be optimized to get increased seed set and yield in female parent of tomato

under protected condition as in shade house. Keeping these points in view, a study has been carried out from the point of seed production, for enhancing the average seed yield and quality of tomato hybrid, under shade house condition.

## MATERIAL AND METHODS

The investigations were carried out on pollen viability of pollen parent (male) and stigma receptivity of seed parent (female) in parental line of Pusa Hybrid-2 tomato under shade house. The shade house experiment was carried out at Hi-Tech Horticulture Unit, Saidapur Farm Main Agriculture Research Station, UAS Dharwad. The laboratory studies were carried out at Seed Quality and Research Laboratory, National Seed Project, UAS Dharwad. The foundation seeds of Pusa hybrid-2 (S-120 x Gavrou) male and female parents were obtained from the Senior Breeder (Tomato), IARI, New Delhi.

The experiment was laid out in female and male block system separately. Female block size was  $5 \times 18 \text{ m}^2$  in which 250 number of female parental plant were transplanted to get sufficient flower buds. In the complete experiment, totally 1200 flower buds were crossed, among which 20 flower buds were crossed with respect to each treatment in each replication and different colour threads were tied to the pedicel of the crossed buds as per the treatment, for easy identification.

Shade house was constructed by use of galvanized iron pipes and covered by 35 per cent green colour shading net. The size of the shade house was  $1280 \text{ m}^2$  ( $40 \times 32 \text{ m}$ ), having 3.50 m height.

Pollen was collected and loaded in pollen rings and were used for pollinating the emasculated flower by dipping the stigma in rings to effect pollination. The buds and flowers that appeared subsequently after the completion of crossing program was manually removed to facilitate better development of the crossed fruits and to avoid selfed fruits in the seed parent. For pollen storage, the pollen collected from male parent were transferred to plastic cup containers with the help of a smooth brush and the cups were stored under ambient room condition where

mean temperature of 24°C and Relative humidity 66 % prevailed. Such stored pollen was used for pollination.

#### Treatment details

Factor-I -Pollen viability of pollen parent (P) – four treatments  $P_0$ : Pollination immediately after collection (fresh pollen)  $P_1$ : One day stored Pollen  $P_2$ : Two days stored Pollen  $P_3$ : Three days stored Pollen Factor-II Stigma receptivity of seed parent (R) – five treatments  $R_0$ : Pollination on the same day of emasculation  $R_1$ : Pollination one day after emasculation  $R_2$ : Pollination two days after emasculation  $R_3$ : Pollination three days after emasculation  $R_4$ : Pollination four days after emasculation. Totally twenty treatment combinations were used in the experiment  $T_1 - P_0R_0, T_2 - P_0R_1, T_3 - P_0R_2, T_4 - P_0R_3, T_5 - P_0R_4, T_6 - P_1R_0, T_7 - P_1R_1, T_8 - P_1R_2, T_9 - P_1R_3, T_{10} - P_1R_4, T_{11} - P_2R_0, T_{12} - P_2R_1, T_{13} - P_2R_2, T_{14} - P_2R_3, T_{15} - P_2R_4, T_{16} - P_3R_0, T_{17} - P_3R_1, T_{18} - P_3R_2, T_{19} - P_3R_3$  and  $T_{20} - P_3R_4$

The data collected in respect to various parameters, on growth and yield attributes, were analyzed statistically as described by [5]. The critical difference (CD) values were calculated at 5 per cent ( $P=0.05$ ) probability level where 'F' test was significant. The data on percentage of germination and seed recovery percentage were transformed into arcsine square root percentage values and transferred data was used for statistical analysis [6].

## RESULTS AND DISCUSSION

The knowledge of pollen viability and stigma receptivity is very important in hybrid seed production, particularly in crop like tomato where the manual emasculation and pollination is involved. The present investigation has helped in providing information on pollen viability and stigma receptivity under protected condition such as shade house, which will enable the protected structure to be adopted by hybrid seed producers.

Collection of pollen and its storage for use in artificial pollination is extremely critical, especially under weather conditions which are not favorable for pollination. Here, the efficiency of

techniques become especially apparent, if pollen need to be transported to far off places and to be utilized where pollen parent growth development is not satisfactory.

#### Pollen parent (P)

The pollen parent exhibited significant differences for various parameters such as fruit set percentage, fruit weight, number of seeds per fruit, seed weight per fruit, seed weight per plant and seed recovery percentage and were significantly higher (67.68%, 95.13g, 114.83, 0.38g, 7.44g and 0.40% respectively) in the two day stored pollen  $P_2$  used for crossing.

This may be attributed to the fact that the two day stored pollen may have more number of mature and viable pollen and this might have resulted in higher fruit setting and seed yield components. Irrespective of stigma receptivity of seed parent, fresh pollen recorded significantly lowest values for fruit setting and seed yield components followed by three days stored pollen, which may be attributed to the immature pollen and shorter pollen viability period respectively, which might have resulted in poor fruit set and seed yield components. These results are in agreement with the findings of [7], [8] and [3] in tomato and [9] in water melon.

Significantly higher values were recorded for seed quality parameters such as germination percentage, root length, shoot length, seed vigour index and seedling dry weight (92.40%, 9.73 cm, 8.0 cm, 1648 and 5.50 mg<sup>-10</sup> respectively) when two day stored pollen ( $P_2$ ) was used for crossing.

Significant increase in the germination percentage, root length, shoot length, seed vigour index and seedling dry weight in two day stored pollen may also have led to the higher seed weight coupled with bolder and well developed seeds obtained from the fruit of two days stored pollen compared to those obtained from fruits of one day stored pollen. These findings are also supported by the work of other research workers like [10] in brinjal and [3] in tomato and [9] in watermelon.

**Table 1.** Effect of pollen viability of pollen parent and stigma receptivity of seed parent on fruits set percentage, fruit weight, number of seeds per fruit, Seed weight per fruit and Seed recovery percentage in hybrid seed production of tomato under shade house

Treat-ments	Fruits set (%)	Fruit weight (g)	No. of seeds/ fruit	Seed weight/ fruit(g)	Seed recovery (%)
<b>Pollen viability of pollen parent (P)</b>					
SP <sub>0</sub>	51.0	83.75	84.07	0.25	0.28(3.03)
P <sub>1</sub>	61.9	89.01	97.13	0.32	0.36(3.43)
P <sub>2</sub>	66.6	92.47	106.47	0.36	0.39(3.56)
P <sub>3</sub>	56.5	84.76	91.15	0.30	0.35(3.37)
SEm±	0.4	0.46	0.65	0.00	0.01(0.03)
CD	1.3	1.31	1.87	0.01	0.02(0.08)
<b>Stigma receptivity of seed parent (R)</b>					
R <sub>0</sub>	43.1	77.03	71.42	0.21	0.27(2.96)
R <sub>1</sub>	61.2	89.96	96.33	0.32	0.35(3.39)
R <sub>2</sub>	67.7	95.13	114.83	0.38	0.40(3.62)
R <sub>3</sub>	65.1	93.60	106.33	0.35	0.37(3.49)
R <sub>4</sub>	58.0	88.02	84.60	0.28	0.32(3.25)
SEm±	0.5	0.51	0.73	0.00	0.01(0.03)
CD	1.4	1.46	2.09	0.01	0.02(0.08)
<b>Interaction (PxR)</b>					
P <sub>0</sub> × R <sub>0</sub>	32.4	71.70	59.33	0.16	0.22(2.71)
P <sub>0</sub> × R <sub>1</sub>	52.1	91.13	87.00	0.27	0.30(3.14)
P <sub>0</sub> × R <sub>2</sub>	62.3	95.30	103.67	0.31	0.33(3.29)
P <sub>0</sub> × R <sub>3</sub>	58.8	94.93	94.67	0.27	0.29(3.07)
P <sub>0</sub> × R <sub>4</sub>	49.6	90.67	75.67	0.24	0.26(2.93)
P <sub>1</sub> × R <sub>0</sub>	46.7	79.57	74.33	0.23	0.29(3.10)
P <sub>1</sub> × R <sub>1</sub>	65.3	89.00	100.33	0.32	0.36(3.42)
P <sub>1</sub> × R <sub>2</sub>	68.2	95.57	116.00	0.39	0.41(3.68)
P <sub>1</sub> × R <sub>3</sub>	66.7	93.83	109.33	0.37	0.39(3.58)

P <sub>1</sub> × R <sub>4</sub>	62.4	87.07	85.67	0.30	0.34(3.36)
P <sub>2</sub> × R <sub>0</sub>	51.4	82.63	82.33	0.25	0.31(3.17)
P <sub>2</sub> × R <sub>1</sub>	69.0	93.30	104.67	0.36	0.39(3.56)
P <sub>2</sub> × R <sub>2</sub>	75.2	98.87	129.00	0.46	0.47(3.91)
P <sub>2</sub> × R <sub>3</sub>	72.2	96.37	121.00	0.42	0.44(3.80)
P <sub>2</sub> × R <sub>4</sub>	65.3	91.20	95.33	0.31	0.34(3.34)
P <sub>3</sub> × R <sub>0</sub>	41.7	74.23	69.67	0.19	0.25(2.87)
P <sub>3</sub> × R <sub>1</sub>	58.6	86.40	93.33	0.31	0.36(3.45)
P <sub>3</sub> × R <sub>2</sub>	65.0	90.77	110.67	0.36	0.40(3.62)
P <sub>3</sub> × R <sub>3</sub>	62.8	89.27	100.33	0.34	0.38(3.52)
P <sub>3</sub> × R <sub>4</sub>	54.6	83.13	81.73	0.29	0.35(3.38)
SEm±	1.0	1.02	1.46	0.01	0.01 (0.06)
CD	2.9	2.93	4.19	0.03	0.03 (NS)

P<sub>0</sub>: Immediately after collection and pollination; P<sub>1</sub>: One day stored Pollen; P<sub>2</sub>: Two day Stored Pollen; P<sub>3</sub>: Three day Stored Pollen; R<sub>0</sub>: Pollination on the same day of emasculatation; R<sub>1</sub>: Pollination one day after emasculatation; R<sub>2</sub>: Pollination two days after emasculatation; R<sub>3</sub>: Pollination three days after emasculatation; R<sub>4</sub>: Pollination four days after emasculatation

#### Seed parent (R)

Among the stigma receptivity studies of seed parent, the treatment R<sub>2</sub> (pollination two days after emasculatation) recorded significantly higher fruit set, fruit weight, number of seeds per fruit, seed weight per fruit, seed weight per plant and seed recovery (67.68%, 95.13g, 114.83, 0.38g, 7.44g and 0.40% respectively), followed by the R<sub>3</sub> treatment. The stigma becomes fully receptive, as revealed by the per cent fruit set, during pollination on second day after emasculatation and thereafter declined.

This suggests that the peak stigma receptivity was attained two days after emasculatation, at Dharwad condition under shade house and on the other hand delayed pollination, particularly pollination four days after emasculatation leads to the lowest fruit set and lesser number of seeds per fruit. This may be due to the drying of stigmatic surface that led to the reduction in fruit

**Table 2.** Effect of pollen viability of pollen parent and stigma receptivity of seed parent on 100 seed weight, germination (%), root length, shoot length, seed vigour index and seedling dry weight in hybrid seed production of tomato under shade house

Treat-ments	100 seed weight (g)	Germination (%)	Root length (cm)	Shoot length (cm)	Seedling vigour index	Seedling dry weight ( $\text{mg}^{-10}$ )
<b>Pollen viability of pollen parent (P)</b>						
P <sub>0</sub>	0.555	86.13(68.4)	9.3	7.4	1441	5.21
P <sub>1</sub>	0.674	91.13(73.0)	9.5	7.9	1587	5.32
P <sub>2</sub>	0.700	92.40(74.6)	9.7	8.0	1648	5.50
P <sub>3</sub>	0.646	88.93(70.9)	9.1	7.4	1477	5.27
<b>SEm±</b>	0.004	0.35(0.3)	0.1	0.1	8	0.05
<b>CD</b>	0.011	1.00(1.0)	0.2	0.2	23	0.14
<b>Stigma receptivity of seed parent (R)</b>						
R <sub>0</sub>	0.558	82.42(65.3)	8.2	6.8	1231	4.87
R <sub>1</sub>	0.656	90.83(72.5)	9.5	7.6	1552	5.21
R <sub>2</sub>	0.704	94.33(76.5)	10.2	8.4	1760	5.86
R <sub>3</sub>	0.683	92.75(74.6)	9.9	8.2	1679	5.70
R <sub>4</sub>	0.620	87.92(69.7)	9.2	7.5	1470	4.99
<b>SEm±</b>	0.004	0.39(0.4)	0.1	0.1	9	0.06
<b>CD</b>	0.012	1.12(1.1)	0.2	0.2	26	0.16
<b>Interaction (P×R)</b>						
P <sub>0</sub> ×R <sub>0</sub>	0.453	77.00(61.3)	7.9	6.4	1105	4.57
P <sub>0</sub> ×R <sub>1</sub>	0.557	87.33(69.1)	9.2	7.2	1435	5.07
P <sub>0</sub> ×R <sub>2</sub>	0.633	91.00(72.5)	10.2	8.2	1671	5.87
P <sub>0</sub> ×R <sub>3</sub>	0.607	89.67(71.2)	10.1	7.9	1614	5.70
P <sub>0</sub> ×R <sub>4</sub>	0.527	85.67(67.7)	9.1	7.0	1379	4.87
P <sub>1</sub> ×R <sub>0</sub>	0.587	85.33(67.5)	8.2	7.0	1297	4.80
P <sub>1</sub> ×R <sub>1</sub>	0.693	92.67(74.3)	9.6	7.9	1628	5.27
P <sub>1</sub> ×R <sub>2</sub>	0.728	95.33(77.5)	10.1	8.5	1773	5.87

P <sub>1</sub> ×R <sub>3</sub>	0.713	93.00(74.3)	9.9	8.3	1693	5.67
P <sub>1</sub> ×R <sub>4</sub>	0.650	89.33(71.0)	9.4	7.9	1542	5.00
P <sub>2</sub> ×R <sub>0</sub>	0.610	86.33(68.3)	8.3	7.1	1332	4.97
P <sub>2</sub> ×R <sub>1</sub>	0.710	92.33(73.9)	9.7	7.9	1622	5.37
P <sub>2</sub> ×R <sub>2</sub>	0.753	97.33(80.7)	10.9	9.0	1934	6.13
P <sub>2</sub> ×R <sub>3</sub>	0.743	96.33(79.0)	10.3	8.7	1830	5.93
P <sub>2</sub> ×R <sub>4</sub>	0.683	89.67(71.2)	9.4	7.6	1524	5.10
P <sub>3</sub> ×R <sub>0</sub>	0.580	81.00(64.1)	8.2	6.5	1191	5.13
P <sub>3</sub> ×R <sub>1</sub>	0.663	91.00(72.5)	9.3	7.4	1523	5.13
P <sub>3</sub> ×R <sub>2</sub>	0.700	93.67(75.4)	9.8	7.9	1661	5.57
P <sub>3</sub> ×R <sub>3</sub>	0.667	92.00(73.6)	9.3	7.8	1579	5.50
P <sub>3</sub> ×R <sub>4</sub>	0.620	87.00(68.8)	9.0	7.5	1433	5.00
<b>SEm±</b>	0.008	0.78(0.8)	0.2	0.1	18	0.11
<b>CD</b>	0.024	2.24	NS	NS	52	NS

P<sub>0</sub>: Immediately after collection and pollination; P<sub>1</sub>: One day stored Pollen; P<sub>2</sub>: Two day Stored Pollen; P<sub>3</sub>: Three day Stored Pollen; R<sub>0</sub>: Pollination on the same day of emasculation; R<sub>1</sub>: Pollination one day after emasculation; R<sub>2</sub>: Pollination two days after emasculation; R<sub>3</sub>: Pollination three days after emasculation; R<sub>4</sub>: Pollination four days after emasculation

set percentage. Pollinating the fresh stigma and immature stigma resulted in reduced fruit set, leading to the lesser number of seed per fruit. These findings are in agreement with the reports of [4], [11] [3] in tomato and [12] in chilli.

The R<sub>2</sub> (pollination two days after emasculation) recorded significantly higher values for seed quality parameters such as germination percentage, root length, shoot length, seed vigour index and seedling dry weight (94.33%, 10.24 cm, 8.4 cm, 1760 and 5.86  $\text{mg}^{-10}$  respectively) followed by the R<sub>3</sub>, as compared to R<sub>2</sub>, R<sub>4</sub> and R<sub>0</sub>.

This may be due to higher seed weight and also bolder seeds that were harvested from this treatment. The increase in seedling vigour index can also be attributed to higher per cent of germination, root length and shoot length, since seedling vigour index is obtained by multiplication of germination percentage with the

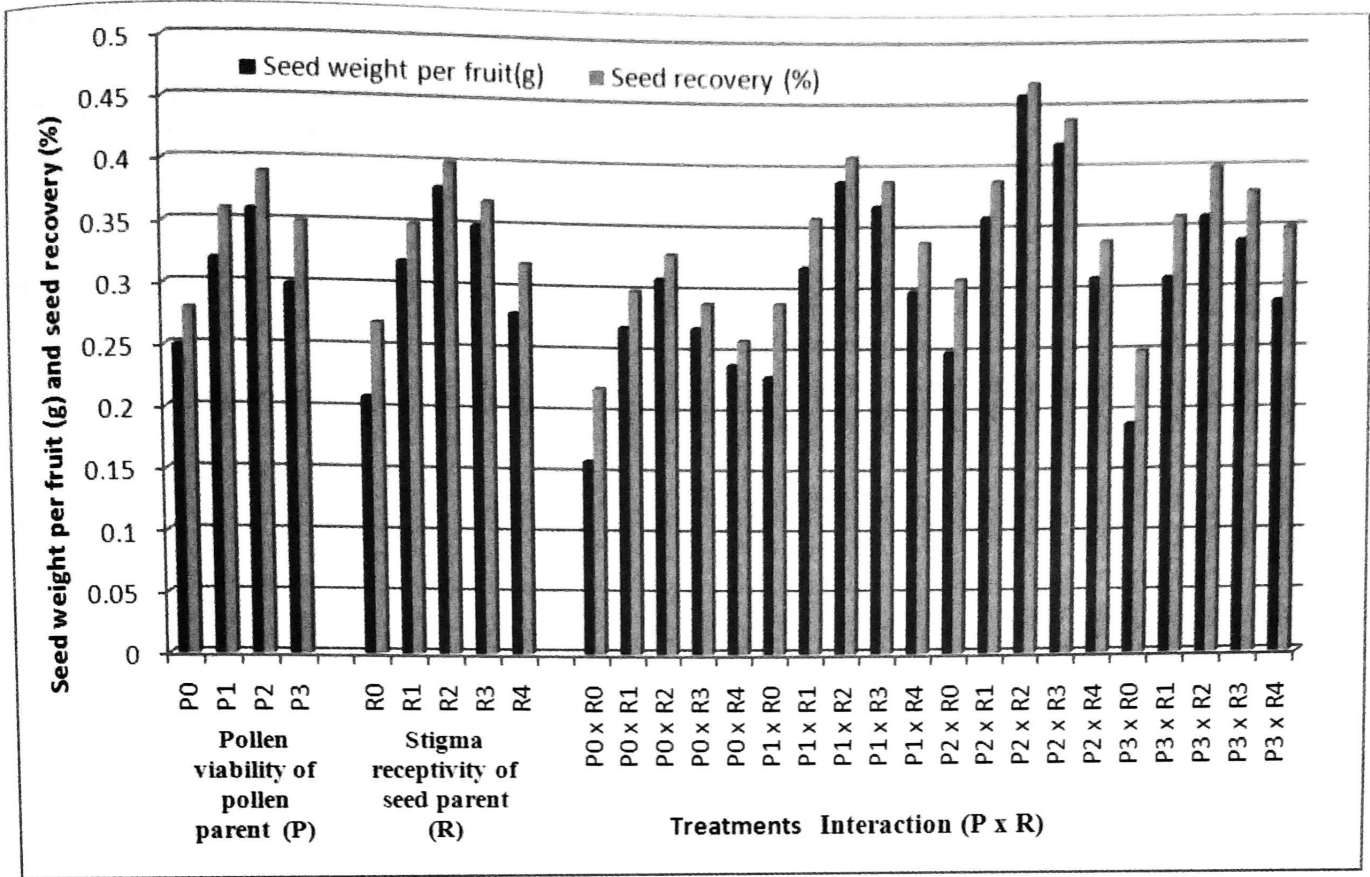


Fig. 1. Effect of pollen viability of pollen parent and stigma receptivity of seed parent on seed weight per fruit and seed recovery percentage in hybrid seed production of tomato under shade house

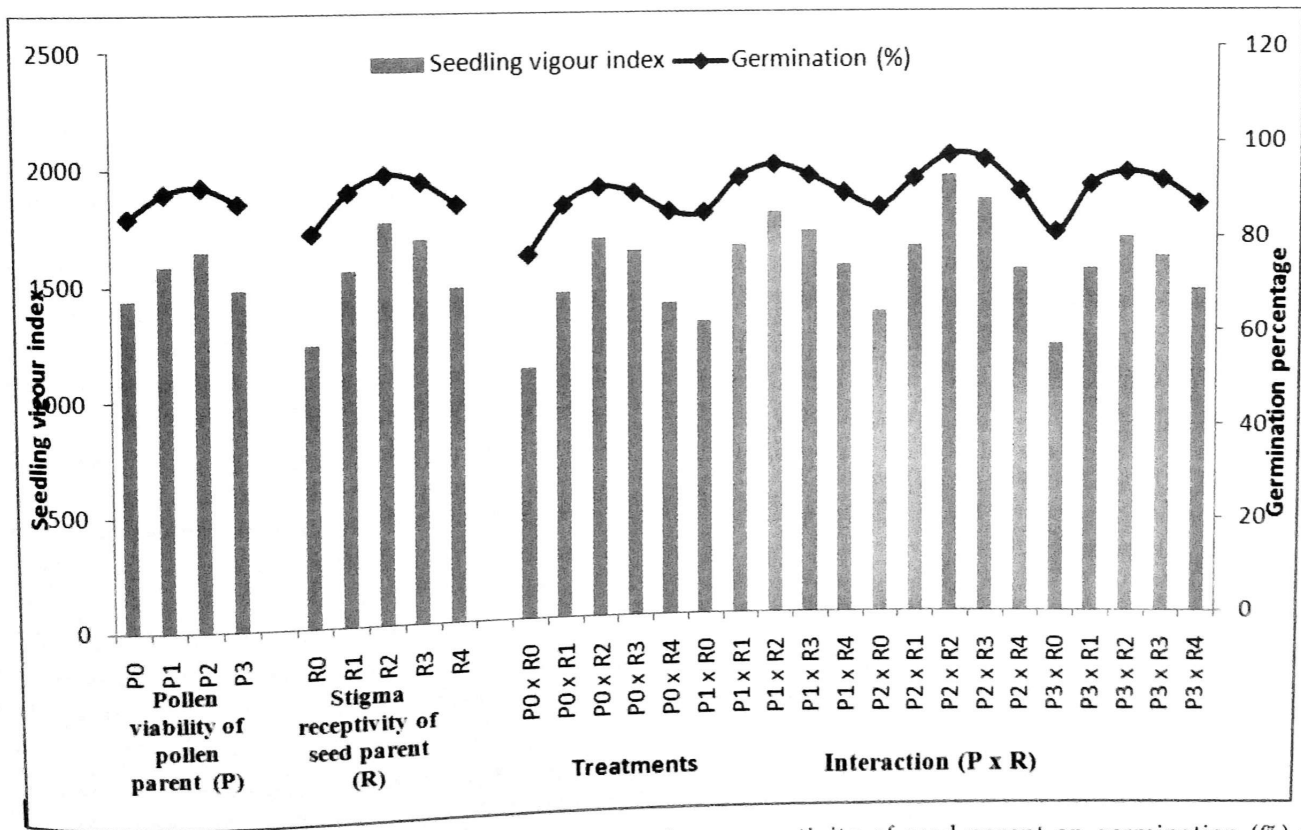


Fig. 2. Effect of pollen viability of pollen parent and stigma receptivity of seed parent on germination (%) and seedling vigour index in hybrid seed production of tomato under shade house

sum total of root and shoot length. Such results were reported earlier by [11], [3] in tomato and [12] in chilli.

#### Interaction effects (PXR)

Among the interaction between pollen viability of pollen parent (P) and stigma receptivity of seed parent (R) (PxR), significantly higher fruit set percentage (75.2%), fruit weight (98.87g), number of seeds per fruit (129), seed weight per fruit (0.46g), seed weight per plant (8.94g), seed recovery percentage (0.47), 100 seed weight (0.753) seed germination percentage (97.33%) and seed vigour index [13] were recorded in the treatment combination of P<sub>2</sub>R<sub>2</sub> (two day stored pollen used for pollination, two days after emasculated Stigma) followed by the combination of P<sub>2</sub>R<sub>3</sub>, as compared to all other combinations.

This may be due to the best combination of mature pollen used for pollinating stigma which was at peak receptivity two days after emasculation, compared to all other treatment combinations. Use of fresh pollen to pollinate fresh stigma soon after emasculation recorded significantly lower (32.39%, 71.70g, 59.33, 0.16 g, 3.11g 0.22%, 77%, 7.93cm, 6.4cm, 1105 and 4.57 mg<sup>-10</sup>, respectively) this is because of poor combination between fresh immature pollen and fresh non-receptive stigma. These findings are in agreement with the reports of Jolli, *et. al*, (2004) in tomato hybrid seed production.

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