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Effect of GA₃ and growing media on seed germination and growth of Custard apple (*Annona squamosa* L.)

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Abstract

An investigation was carried out to see the effect of GA₃ and growing media on seed germination and growth of Custard apple (*Annona squamosa* L.) at Fruit Research Station, Imalia Farm, Department of Horticulture, College of Agriculture, JNKVV, Jabalpur (M.P.) Amongst different growing media, all germination parameters like days taken to start germination, days taken to 50% germination, percent of germination in each treatment at 60 DAS and almost growth parameters like height of shoots, number of leaves per seedling, height of seedling, root length, number of roots/seedling, fresh weight of shoots, dry weight of shoots, fresh weight of roots, dry weight of roots were found significantly superior under M₅ growing media comprising soil + vermicompost + AZO + PSB followed by M₂ (soil + vermicompost), M₄ (soil + FYM + AZO + PSB), M₁ (soil + FYM), M₃ (soil + AZO + PSB), M₀ (only soil) at 60, 90, 120 and 150 days after sowing, respectively. But mean value of girth of stem was observed slightly better in the treatments having FYM and bio-fertilizers (M₄) closely followed by M₁ (soil + FYM) and M₅ (Soil + Vermicompost + Azotobacter + PSB). The concentration of 600 ppm (G₃) was observed superior in germination point of view like days taken to start germination, days taken to 50% germination, percent of germination in each treatment and found statistically at par with other concentration of GA₃ except G₀ (water soaked seed). As regards the interactions, all germination parameters and almost growth parameters like height of shoots, number of leaves per seedling, girth of stem, height of seedling, root length, number of roots/seedling, fresh weight of shoots, dry weight of shoots, fresh weight of roots, dry weight of roots were found not significant. However, G₂M₅ treatment combination [seed soaked in 400 ppm GA and sown in soil + vermicompost + AZO + PSB] recorded superior and G₀M₀ combination (control) was found inferior in respect to survival and most of growth parameters. G₃M₅ treatment combination [seeds soaked in 600 ppm GA concentration and sown in soil + vermicompost + AZO + PSB] showed superiority over rest of the treatment combinations with respect to germination whereas minimum was observed in control (G₀M₀).

Keywords: Germination, GA₃, Biofertilizers, Growing media, Growth

1. Introduction

Custard apple (*Annona squamosa* L.) belongs to family Annonaceae and is one of the finest fruits gifted to India by tropical America. It is commonly found in India and cultivated an area of 23 thousand ha with production of 176 MT (Anonymous, 2013)^[1]. Custard apple, popularly known as Sitaphal is grown mainly in the States of Andhra Pradesh, Assam, Tamilnadu, Madhya Pradesh and grows wild in Deccan plateau and some parts of central India. Custard apple is generally classified as semi wild fruit by virtue of its spontaneous spread in forests, wastelands and other uncultivated places. It is hardy, tolerant to drought, salinity and saline irrigation water to certain extent. It grows very well even on a shallow soil. It also sheds off leaves during stress period to minimize the moisture loss from plant tissues through transpiration and thus a most appropriate fruit crop for rainfed region. Custard apple is known by varied name like Sitaphal, Sugar apple and Shariffa. It is considered as one of the delicious and nutritionally valuable fruits meant for table purpose. Fruits have an edible, soft, granular, juicy and sugary pulp with mild flavour and with slight acidity. Fruits are considered for their medicinal value besides their general use in ice cream, confectionery, certain milk products and in making preserves as jam, jelly and other products. It is considered as beneficial for cardiac disease, diabetes, hyperthyroidism and cancer. It contains about 28-55% of edible portion consisting of 73.30% moisture, 1.60% protein, 0.30% fat, 0.70% mineral matter, 23.90% carbohydrates, 0.20% calcium, 0.40% phosphorus,

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1.0% iron, 12.4-18.15% sugar and 0.26-0.65% acidity with caloric value of 105 K Cal/100g. It is generally propagated by seed since there is little variability among seedlings. Maximum germination can be obtained by sowing of freshly extracted seed upto 20-30 days. The seeds of Annonaceae are albuminous ellipsoids and their length varies between 5 and 30 mm. They have a ruminate endosperm (Corner, 1976) [4]. The embryo is small, straight, with moderately developed embryonic axis, rudimentary plumule and a flat and thin cotyledon; which develops after the seed is formed (Corner, 1976) [4]. Setten and Koek-Noorman (1992) [20] observed that Annonaceae seeds undergoing dispersal have a small embryo that is considered underdeveloped and immature; immaturity requires time to complete embryo growth after seed dispersion. Meanwhile, Hayat (1963) [5] reported that the seeds of *A. squamosa* have a small embryo with two foliaceous, thin cotyledons that take one to three months to germinate.

Seed germination is the resumption of active growth of embryo that results in the emergence of the young plant. Seeds of many fruit crops remain ungerminated even under favourable conditions. Such kind of dormancy in seeds may be due to presence of hard and impermeable seedcoat, germination inhibitors and improper development of embryo. Such seeds may require special treatments like scarification, soaking in water, growth regulators etc. to overcome dormancy. Gibberellins (GA_3) activate the embryonic vegetative growth, weakens the endosperm layer that involves the embryo and restricts its growth, and mobilizes the energetic reserves from the endosperm of cereals (Bewley, 1997^[2]; Taiz and Zeiger, 2006 ^[24]). Cereal embryo synthesizes and releases GA during the germination, which leads to the production and/or secretion of several hydrolytic enzymes involved in the solubilization of reserves, including α -amylase and β -amylase (Taiz and Zeiger, 2006) ^[24]. Growing media is one of the important environmental factors, which plays an important role in growth and survival of seedlings. Several growing media or their combinations are being used for raising the seedling. Different growing media like soil, sand, farm yard manure (FYM) and vermicompost either alone or in different proportion have been found

beneficial to influence germination and growth of seedling. A good growing media provides sufficient anchorage or support to the plant, serves as a reservoir for nutrients and water, allows oxygen diffusion to the roots and permits gaseous exchange between roots and the atmosphere outside root substrate. Bio-fertilizers are also beneficial in seed germination and growth of seedling. Bio-fertilizers are the carrier-based preparations containing mainly effective strains of microorganisms in sufficient number, which are useful for nitrogen fixation. Amongst bio-fertilizers *Azotobacter* strains play a key role in harnessing the atmospheric nitrogen through its fixation in the roots. They have been also reported to improve fertility condition of the soil. The seed coat of most of the fruit crops is very hard. To break the seed dormancy, either some chemical treatment or long incubation period is required but bio-inoculants like *Azotobacter*, PSB etc. also can be helpful in breaking the seed dormancy by producing various plant growth substances in combination with either farmyard manure (FYM) or vermicompost on seed germination and plant growth.

2. Materials and methods

The experiment was carried out at Fruit Research Station, Imalia, JNKVV, Jabalpur during the year 2014-2015. Jabalpur is situated in "Kymore Plateau and Satpura Hills" Agro-climatic zone of Madhya Pradesh at 23.9°N latitude and 79.58° east longitudes and an altitude of 411.78 meters above the mean sea level. The tropic of cancer passes through the middle of the district. The climate of Jabalpur region is semi-arid and subtropical having warm and dry spring summer and cool winter as main characteristics feature, in general the highest temperature reaches above 45°C and below 5°C respectively. The relative humidity varies from 70-80%. The average annual rainfall is about 1375, mm, which is mainly distributed from mid June to first week of October from south-west monsoon with occasional rain during winter. The experiment was laid out with Factorial RBD along with six different growing media and four different concentration of Gibberellic acid with three replications.

2.1 Treatment details

Treatments	Treatment combinations	composition
T ₁	G ₀ M ₀	0 ppm GA ₃ + Soil (Control)
T ₂	G ₀ M ₁	0 ppm GA ₃ + (Soil + FYM)
T ₃	G ₀ M ₂	0 ppm GA ₃ + (Soil + Vermicompost)
T ₄	G ₀ M ₃	0 ppm GA ₃ + (Soil + AZO + PSB)
T ₅	G ₀ M ₄	0 ppm GA ₃ + (Soil + FYM + AZO + PSB)
T ₆	G ₀ M ₅	0 ppm GA ₃ + (soil + Vermicompost + AZO + PSB)
T ₇	G ₁ M ₀	200 ppm GA ₃ + Soil
T ₈	G ₁ M ₁	200 ppm GA ₃ + (Soil + FYM)
T ₉	G ₁ M ₂	200 ppm GA ₃ + (Soil + Vermicompost)
T ₁₀	G ₁ M ₃	200 ppm GA ₃ + (Soil + AZO + PSB)
T ₁₁	G ₁ M ₄	200 ppm GA ₃ + (Soil + FYM + AZO + PSB)
T ₁₂	G ₁ M ₅	200 ppm GA ₃ + (soil + Vermicompost + AZO + PSB)
T ₁₃	G ₂ M ₀	400 ppm GA ₃ + Soil
T ₁₄	G ₂ M ₁	400 ppm GA ₃ + (soil + FYM)
T ₁₅	G ₂ M ₂	400 ppm GA ₃ + (Soil + Vermicompost)
T ₁₆	G ₂ M ₃	400 ppm GA ₃ + (Soil + AZO + PSB)
T ₁₇	G ₂ M ₄	400 ppm GA ₃ + (Soil + FYM + AZO + PSB)
T ₁₈	G ₂ M ₅	400 ppm GA ₃ + (soil + Vermicompost + AZO + PSB)
T ₁₉	G ₃ M ₀	600 ppm GA ₃ + Soil
T ₂₀	G ₃ M ₁	600 ppm GA ₃ + (Soil + FYM)
T ₂₁	G ₃ M ₂	600 ppm GA ₃ + (Soil + Vermicompost)
T ₂₂	G ₃ M ₃	600 ppm GA ₃ + (Soil + AZO + PSB)
T ₂₃	G ₃ M ₄	600 ppm GA ₃ + (Soil + FYM + AZO + PSB)
T ₂₄	G ₃ M ₅	600 ppm GA ₃ + (soil + Vermicompost + AZO + PSB)

2.2 Preparation of growing media

- The media were used to grow seedling in polybags comprising of
1. Only soil
 2. Soil + FYM – the ratio of 2:1 of soil and fully decomposed FYM.
 3. Soil + Vermicompost – the ratio of 2:1 of soil and vermicompost.
 4. Soil + AZO + PSB – soil enrich with AZO and PSB (each 5 g/kg soil).
 5. Soil + FYM + AZO + PSB – the ratio of 2:1 of soil and FYM enrich with AZO and PSB (each 5 g/kg soil).
 6. Soil + Vermicompost + AZO + PSB – the ratio of 2:1 of soil and vermicompost enrich with AZO and PSB (each 5 g/kg soil).

2.3 Germination percentage

Days taken to start germination, 50 per cent germination and germination in each treatment were recorded at 60 days after sowing. Number of seedlings were counted and expressed as germination percentage.

$$\text{Germination (\%)} = \frac{\text{Total no. of seeds germinated}}{\text{Total no of seeds sown}} \times 100$$

2.4 Height of shoot (cm)

The length from the collar region to the tip of the shoot apex was measured for five randomly selected tagged plants in each treatment at 30, 60, 90, 120 and 150 days after sowing and the average of length was computed.

2.5 Girth of stem (mm)

The diameter of shoot above the root collar region was recorded using vernier caliper.

2.6 Length of seedling (cm)

The height of seedling is measured from root tip to the shoot tip and expressed in centimeter at 150 days after sowing.

2.7 Root length (cm)

The length from collar region to the tip of the root was measured for five randomly uprooted plants in each treatment. The average value was computed.

2.8 Number of roots per seedling

In number of roots per seedling, primary, secondary and tertiary and rootlets were counted.

2.9 Fresh weight of seedling

The plants were carefully washed to remove the soil adhering to their roots and shoots. The weight was taken with the help of electronic balance and average value was computed.

2.1 Dry weight of seedling

For dry weight plant were chopped and oven dried at $60 \pm 2^\circ\text{C}$ temperature till a constant weight. The weight was taken with the help of electronic balance and average value was computed.

3. Results and Discussions

3.1 Days taken to start germination

It is obvious from the present research that days taken to start germination was significantly minimum (31.50) under growing media M₅ having soil + vermicompost + AZO +

PSB. Days taken to start germination were slightly better in the treatments having vermicompost over their respective FYM treatments. It might be due to variation in characteristic of different media. Medium M₅ has relatively high content of humus-like compounds, active micro organisms, enzymes as well as physical and nutritional condition of media which increased the physiological activities of seed and initiate the early germination. The finding is similar to Sahni *et al.* (2008)^[19] and Vasu *et al.* (2010)^[25] who reported that 10 g inoculation of *Azotobacter* showed 100% germination and also reduced the average time taken to start germination. Gibberellic acid concentration also significantly affected initiation of seed germination. The minimum days (32.61 days) taken to initiate the germination was under G₃ (600 ppm) while the maximum days (35.44 days) under control (G₀) when seed soaking in water alone. This increment in germination is due to exogenous application of Gibberellic acid which antagonizes the ill effect of inhibitors along with increase in cell wall plasticity and better water absorption. The findings are supported by Ratan and Reddy (2004)^[17] who reported that the GA₃ 600 ppm took minimum time to germinate the seeds of Custard apple. The interaction of growing media and gibberellic acid showed non significant effect on initiation of seed germination and the minimum days (30.00 days) taken to initiate germination were recorded under the treatment combination G₃M₅ Comprising soil + vermicompost + AZO + PSB + 600 ppm GA₃ as compared to other combinations of growing media and concentration of GA₃. It is probably due to the synergistic combination of both the factors. The finding was supported by Jain and Parmar (1993)^[6].

3.2 Days taken to 50% germination

The present research reveals that the growing media and GA₃ showed significant effect on germination. The minimum days was taken to 50 % germination (46.83 days) under the growing media M₅ and maximum (51.42 days) taken to 50 % germination under the media M₀. Initiation of seed germination was significantly affected by gibberellic acid. The minimum days (47.17 days) taken to 50% germination under G₃ (600 ppm) while the maximum days (51.06 days) taken to under G₀. The findings are supported by Ynoue *et al.* (1999)^[27] who reported that the GA₃ 150 ppm reduced the average time of germination on kiwi fruit seeds. The interaction effect of growing media and gibberellic acid found to be non-significant on 50% seed germination, however, the minimum days (44.33 days) taken to 50% germination were noted under the treatment combination of G₃M₅. It might be due to synergistic effect of both factor help to early initiation of seed germination. The finding was supported by Jain and Parmar (1993)^[6].

3.3 Percentage of germination at 60 DAS

The highest germination percentage (77.50%) was obtained under the media M₅ comprising soil + vermicompost + AZO + PSB. This finding is supported by Sinish *et al.* (2005)^[22] who reported that combined inoculation of AZO, PSB and AMF in the potting mixture induced better germination percentage. Gibberellic acid concentration showed significant effect on percentage of germination at 60 DAS. The maximum germination percentage of 73.89 was recorded under G₃ (600 ppm GA) and the minimum germination (59.44%) was recorded under G₀. It might be due to GA₃ which accelerate the activity of specific enzymes such as α -amylase, which have brought an increase in availability of starch assimilation

resulting an early germination. Interaction effect of growing media and gibberellic acid did not show significant effect on seed germination percentage whereas maximum germination percentage (86.67%) was noted under the combination of growing media and gibberellic acid G₃M₅ (soil + vermicompost + AZO + PSB + 600 ppm). It may be due to synergistic effect of both factors.

3.4 Length of seedling (cm)

The maximum height of seedling (43.81cm) at 150 DAS was recorded under the treatment M₅ comprising soil + vermicompost + AZO + PSB. The present investigations are in conformity with the results of Shristava and Bhel (2002)^[21], Sinish *et al.* (2005)^[22], Sahni *et al.* (2008)^[19]. Probable reasons may be that the media M₅ creates sufficient porous space to let the excess water drain away and pertaining

adequate aeration for the better seedling growth. The excellent plant growth in vermicompost was possibly due to some plant growth promoters in worm casts which increase physiological activities of seed, essential for cell division and cell enlargement. Krishnamoorthy and Vajranrabhaian (1986)^[10] who reported that some plant growth promoters such as cytokinins and auxins are present in worm cast. It also contains rich source of nutrient than FYM. The results obtained were closely related with the finding of Kumar and Sharma (2007)^[11] in *Jatropha curcus*. As regard to GA₃, a significant increase in height of seedlings (42.13 cm) at 150 days after sowing was observed. Gibberellins are well known for inter-nodal cell elongation, thereby leading to increase in seedling length. These findings are supported by Ratan and Reddy (2004)^[17].

Table 1: Effect of growing media (M) and gibberellic acid (G) on days taken to start germination (days), percentage of germination and length of seedling (cm)

Treatments	Days taken to start germination	Days taken to 50 % germination	% of germination at 60 DAS	Length of seedling at 150 DAS (cm)
Gibberellic acid				
G ₀ (0 ppm)	35.44	51.06	59.44	37.47
G ₁ (200 ppm)	33.72	48.94	68.89	39.18
G ₂ (400 ppm)	33.11	47.83	72.22	42.13
G ₃ (600 ppm)	32.61	47.17	73.89	40.43
SEm	0.526	0.600	2.604	0.471
CD at 5% level	1.502	1.714	7.436	1.344
Media				
M ₀ (Soil)	36.00	51.42	60.00	34.46
M ₁ (Soil+FYM)	33.92	48.92	66.67	39.96
M ₂ (Soil+VC)	32.67	47.75	72.50	42.85
M ₃ (Soil+BF)	35.08	49.67	64.17	36.16
M ₄ (Soil+FYM+BF)	33.17	46.33	70.83	41.58
M ₅ (Soil+VC+BF)	31.50	44.33	77.50	43.81
SEm	0.644	0.735	3.189	0.576
CD at 5% level	1.840	2.100	9.107	1.646
Interaction				
G ₀ M ₀	37.33	53.00	53.33	31.68
G ₀ M ₁	35.67	52.67	56.67	38.06
G ₀ M ₂	33.67	50.00	60.00	41.28
G ₀ M ₃	37.00	51.67	56.67	33.30
G ₀ M ₄	34.67	49.00	63.33	39.31
G ₀ M ₅	34.33	50.00	66.67	41.18
G ₁ M ₀	36.33	51.67	63.33	32.90
G ₁ M ₁	33.67	48.33	66.67	39.35
G ₁ M ₂	32.67	48.00	73.33	41.77
G ₁ M ₃	35.67	50.33	70.00	36.55
G ₁ M ₄	33.33	48.33	66.67	42.00
G ₁ M ₅	30.67	47.00	73.33	42.54
G ₂ M ₀	35.67	50.67	60.00	37.82
G ₂ M ₁	33.33	49.67	70.00	41.76
G ₂ M ₂	32.33	47.00	76.67	44.87
G ₂ M ₃	33.67	47.67	70.00	38.29
G ₂ M ₄	32.67	48.00	73.33	43.12
G ₂ M ₅	31.00	46.00	83.33	46.90
G ₃ M ₀	34.67	50.33	63.33	35.43
G ₃ M ₁	33.00	47.00	73.33	40.67
G ₃ M ₂	32.00	46.00	80.00	43.67
G ₃ M ₃	34.00	49.00	60.00	36.51
G ₃ M ₄	32.00	46.33	80.00	41.91
G ₃ M ₅	30.00	44.33	86.67	44.61
SEm	1.289	1.471	6.378	1.153
CD at 5% level	NS	NS	NS	NS

3.5 Height of shoot (cm)

Growing media had showed significant effect on maximum height of shoots (i.e. 4.09, 8.33, 12.06 and 17.68 cm),

respectively at 60, 90, 120 and 150 DAS under the treatment M₅. The results are in conformity with the results of Shristava and Bhel (2002)^[21], Sinish *et al.* (2005)^[22] and Kumar and

Sharma (2007) [11]. Growing media comprising soil + vermicompost + AZO + PSB (M_5) creates sufficient porous space to let the excess water drain away and pertaining adequate aeration for the better seedling growth, it may also be helped to increase physiological activities of seed which is essential for cell division, cell elongation resulting more nutrients absorption from the soil media. The results obtained were closely related with the finding of Kumar and Sharma (2007) [11] in *Jatropha curcus*. As regards the gibberellic acid concentration, maximum height of shoots (3.97, 7.65, 11.83 and 17.60 cm) was recorded under G_2 (400 ppm) at 60, 90, 120 and 150 DAS, respectively. Basically, plant height is a genetically controlled character but several studies have indicated that plant height can be increased by application of

synthetic plant growth regulators. However, in the present investigation a significant difference in plant height was noticed by the application of different concentration of GA_3 . It might be due to GA_3 effect on elongation of internodes, as GA_3 is known to enhance cell elongation (Krishnamoorthy and Sandooja, 1981) [9]. Increase in plant height has also been reported by Ratan and Reddy (2004) [17]. Interaction effect of growing media and gibberellic acid shows non-significant and maximum height of shoot at successive growth period was observed under treatment combination G_2M_5 i.e. 4.61, 8.89, 12.97 and 18.63 cm at 60, 90, 120 and 150 days after sowing, respectively whereas minimum height was recorded under G_0M_0 .

Table 2: Influence of growing media and gibberellic acid on height of shoots (cm)

Treatment	60 DAS	90 DAS	120 DAS	150 DAS
Gibberellic acid				
G_0 (0 ppm)	3.09	6.87	10.68	16.02
G_1 (200 ppm)	3.44	7.22	11.16	16.45
G_2 (400 ppm)	3.97	7.65	11.83	17.60
G_3 (600 ppm)	3.75	7.34	11.47	17.05
SEm	0.056	0.104	0.078	0.180
CD at 5% level	0.160	0.298	0.222	0.515
Media				
M_0 (Soil)	2.91	6.23	10.32	15.63
M_1 (Soil+FYM)	3.56	7.13	11.20	16.86
M_2 (Soil+VC)	3.96	8.01	11.82	17.54
M_3 (Soil+BF)	3.10	6.55	10.77	15.96
M_4 (Soil+FYM+BF)	3.76	7.39	11.56	17.03
M_5 (Soil+VC+BF)	4.09	8.33	12.06	17.68
SEm	0.069	0.128	0.095	0.221
CD at 5% level	0.196	0.365	0.272	0.630
Interaction				
G_0M_0	2.47	5.81	9.48	14.33
G_0M_1	3.32	7.17	10.76	16.49
G_0M_2	3.37	7.34	11.19	16.85
G_0M_3	2.57	6.03	10.19	14.69
G_0M_4	3.17	6.97	10.90	16.87
G_0M_5	3.63	7.91	11.56	16.91
G_1M_0	2.56	5.99	10.00	14.52
G_1M_1	3.49	7.07	11.13	16.70
G_1M_2	3.80	8.03	11.71	17.37
G_1M_3	3.19	6.56	10.74	16.03
G_1M_4	3.83	7.49	11.83	16.87
G_1M_5	3.79	8.19	11.56	17.22
G_2M_0	3.42	7.13	10.92	17.20
G_2M_1	3.72	6.92	11.43	17.24
G_2M_2	4.37	8.27	12.43	18.21
G_2M_3	3.53	7.09	11.24	16.91
G_2M_4	4.17	7.61	11.99	17.43
G_2M_5	4.61	8.89	12.97	18.63
G_3M_0	3.20	5.98	10.86	16.44
G_3M_1	3.71	7.36	11.49	17.00
G_3M_2	4.29	8.39	11.93	17.72
G_3M_3	3.12	6.51	10.91	16.22
G_3M_4	3.87	7.48	11.51	16.94
G_3M_5	4.32	8.35	12.13	17.97
SEm	0.137	0.256	0.191	0.441
CD at 5% level	NS	NS	NS	NS

3.6 Number of leaves per seedling

The number of leaves per seedling, the maximum mean number of leaves per seedling was observed under treatment M_5 (4.78, 9.05, 16.10 and 21.32) at 60, 90, 120 and 150 days after sowing, respectively. The improvement in vegetative character might be due to the ability of *Azotobacter* to fix

atmospheric nitrogen which may share its role in increasing the percentage of mineral nutrient in soil. In addition, it increases the surface area of root hairs followed by increase in average absorption of mineral nutrients and ready vermicompost relatively contains more exchangeable plant nutrient especially N_2 (responsible for vegetative growth) than

FYM. The results of Kumar and Sharma (2007)^[11] in *Jatropha curcus*, Krishna *et al.* (2008)^[8] and Peng *et al.* (2013)^[14] have close conformity with present findings. As regards the gibberellic acid concentration, GA₃ showed significant effect on number of leaves and maximum number of leaves per seedling (4.71, 9.00, 16.10 and 21.06) was recorded under G₂. The production of more number of leaves in GA₃ treatments may be due to the vigorous growth and more number of branches induced by GA₃ facilitates better harvest of sunshine

by the plants to produce more number of leaves. Similar findings were also reported by Rao and Reddy (2005)^[16] in mango. The interaction effect of growing media and gibberellic acid showed non significant effect at 60, 90, 120 and 150 days after sowing. However, maximum number of leaves per seedling was recorded in G₂M₅ (5.33, 9.73, 16.93 and 22.07 at 60, 90, 120 and 150 days after sowing, respectively). It may be due to synergistic effect of both factors.

Table 3: Influence of growing media and gibberellic acid on number of leaves per seedling

Treatment	60 DAS	90 DAS	120 DAS	150 DAS
Gibberellic acid				
G ₀ (0 ppm)	3.37	7.58	14.51	19.60
G ₁ (200 ppm)	3.80	8.00	14.96	20.02
G ₂ (400 ppm)	4.71	9.00	16.10	21.06
G ₃ (600 ppm)	4.31	8.53	15.34	20.53
SEm	0.118	0.117	0.129	0.156
CD at 5% level	0.338	0.335	0.369	0.444
Media				
M ₀ (Soil)	3.17	7.38	14.47	19.25
M ₁ (Soil+FYM)	4.02	8.22	15.00	20.20
M ₂ (Soil+VC)	4.45	8.72	15.58	20.78
M ₃ (Soil+BF)	3.68	7.87	14.85	19.87
M ₄ (Soil+FYM+BF)	4.18	8.43	15.37	20.40
M ₅ (Soil+VC+BF)	4.78	9.05	16.10	21.32
SEm	0.145	0.144	0.158	0.191
CD at 5% level	0.414	0.410	0.452	0.544
Interaction				
G ₀ M ₀	2.53	6.73	13.60	18.53
G ₀ M ₁	3.47	7.67	14.47	19.53
G ₀ M ₂	3.73	7.93	14.87	20.07
G ₀ M ₃	2.60	6.80	13.93	19.00
G ₀ M ₄	3.60	7.80	14.80	19.87
G ₀ M ₅	4.27	8.53	15.40	20.60
G ₁ M ₀	2.87	7.07	14.20	19.07
G ₁ M ₁	4.07	8.27	15.27	20.40
G ₁ M ₂	4.07	8.27	15.07	20.13
G ₁ M ₃	3.27	7.47	14.40	19.40
G ₁ M ₄	4.27	8.47	15.47	20.53
G ₁ M ₅	4.27	8.47	15.33	20.60
G ₂ M ₀	3.33	7.60	15.20	19.80
G ₂ M ₁	4.53	8.73	15.87	20.80
G ₂ M ₂	5.00	9.47	16.13	21.27
G ₂ M ₃	4.93	9.07	16.20	21.20
G ₂ M ₄	5.13	9.40	16.27	21.20
G ₂ M ₅	5.33	9.73	16.93	22.07
G ₃ M ₀	3.93	8.13	14.87	19.60
G ₃ M ₁	4.00	8.20	14.40	20.07
G ₃ M ₂	5.00	9.20	16.27	21.67
G ₃ M ₃	3.93	8.13	14.87	19.87
G ₃ M ₄	3.73	8.07	14.93	20.00
G ₃ M ₅	5.27	9.47	16.73	22.00
SEm	0.290	0.287	0.317	0.381
CD at 5% level	NS	NS	NS	NS

3.7 Girth of stem (mm)

The maximum girth of stem (2.71, 3.13, 3.53 and 4.16 mm) was recorded at 60, 90, 120 and 150 DAS in M₄ (soil + FYM + AZO + PSB) whereas minimum girth under treatment M₀. Girth of stem was slightly better in the treatments having FYM over their respective vermicompost treatments. The probable reason may be that the medium M₄ has high anchorage or support to the plant, water retention capacity, allows oxygen diffusion to the roots and permits gaseous

exchange between roots and the atmosphere outside root substrate. Similar findings were also reported by Taiwo (2004)^[23] and Parasana *et al.* (2012)^[12]. As regards to GA₃, the significantly maximum girth of stem (2.57, 2.98, 3.29 and 3.89 mm) was recorded under G₁ and minimum girth of stem was recorded under G₀. The increase in seedling girth by application of gibberellic acid was also reported by Rao and Reddy (2005)^[17] in mango.

Table 4: Influence of growing media and gibberellic acid on girth of stem (mm)

Treatment	60 DAS	90 DAS	120 DAS	150 DAS
Gibberellic acid				
G ₀ (0 ppm)	1.94	2.62	2.92	3.51
G ₁ (200 ppm)	2.57	2.98	3.29	3.89
G ₂ (400 ppm)	2.34	2.92	3.25	3.80
G ₃ (600 ppm)	2.27	2.80	3.14	3.62
SEm	0.095	0.093	0.095	0.101
CD at 5% level	0.272	0.264	0.272	0.289
Media				
M ₀ (Soil)	1.78	2.33	2.72	3.27
M ₁ (Soil+FYM)	2.65	3.09	3.37	3.82
M ₂ (Soil+VC)	2.16	2.88	3.08	3.65
M ₃ (Soil+BF)	2.00	2.64	2.99	3.54
M ₄ (Soil+FYM+BF)	2.71	3.13	3.53	4.16
M ₅ (Soil+VC+BF)	2.40	2.92	3.22	3.80
SEm	0.117	0.113	0.117	0.124
CD at 5% level	0.334	0.324	0.333	0.354
Interaction				
G ₀ M ₀	1.12	2.06	2.48	2.79
G ₀ M ₁	2.03	3.00	3.16	4.02
G ₀ M ₂	1.96	2.66	2.77	3.34
G ₀ M ₃	2.00	2.34	2.82	3.76
G ₀ M ₄	2.41	2.88	3.21	3.48
G ₀ M ₅	2.15	2.77	3.09	3.46
G ₁ M ₀	2.00	2.28	2.83	3.67
G ₁ M ₁	3.08	3.40	3.46	3.81
G ₁ M ₂	2.55	2.95	3.05	3.55
G ₁ M ₃	2.10	2.85	3.09	3.53
G ₁ M ₄	3.18	3.51	4.13	4.71
G ₁ M ₅	2.52	2.92	3.18	3.93
G ₂ M ₀	1.92	2.64	2.77	3.12
G ₂ M ₁	3.05	2.91	3.51	3.57
G ₂ M ₂	2.13	2.88	3.22	4.11
G ₂ M ₃	1.80	2.70	3.05	3.49
G ₂ M ₄	3.00	3.40	3.76	4.54
G ₂ M ₅	2.15	3.01	3.21	3.97
G ₃ M ₀	2.06	2.32	2.81	3.25
G ₃ M ₁	2.45	3.03	3.36	3.86
G ₃ M ₂	2.01	3.01	3.28	3.58
G ₃ M ₃	2.12	2.65	3.01	3.39
G ₃ M ₄	2.25	2.74	2.99	3.92
G ₃ M ₅	2.76	3.01	3.38	3.60
SEm	0.234	0.227	0.233	0.248
CD at 5% level	NS	NS	NS	NS

3.8 Root length (cm)

The maximum root length of 26.13 cm was obtained under treatment M₅ at 150 days after sowing whereas the minimum root length was obtained under treatment M₀ (18.76 cm). It might be due to media having soil + vermicompost + AZO + PSB that create sufficient porous space to permit adequate aeration, water holding capacity, improved soil texture and structure, maintained soil temperature and improved soil health and nutrient status of medium for better root growth. Application of vermicompost enhances the activity of some microbial populations which increase the level of N. This was due to higher N fixer in experimental plot than control. The findings are agreement with the findings of Pathak *et al.* (2009)^[13]. As regards the GA₃, Maximum root length of 24.52 cm was recorded under G₂ which was significantly superior over rest of the treatments. Exogenous application of GA₃ induced the activity of gluconeogenic enzymes during early stages of seed germination and this could be the reason for improved germination and vigour characteristics that is reflected in terms of increase in root length. Similar findings were also reported by Wagh *et al.* (1998)^[26].

3.9 Number of roots per seedling

At 150 days after sowing, the maximum number of roots per seedling (50.37) was observed under M₅ whereas treatments M₀ was recorded minimum. The excellent plant growth in vermicompost was possibly due to some plant growth promoters in worm casts. This improvement of vegetative growth might be due to the role of vermicompost in improving the soil's physiochemical structure, also promotes biological properties of soil, increasing soil ventilation by increasing the porosity and a big source of nutrient elements especially nitrogen and phosphorus. The results of Chopde *et al.* (1999)^[3] are close conformity with present findings. The effect of GA₃ had significant effect on number of roots/seedling. Significantly maximum number of roots (48.90) was noted under G₂. Vigorous root growth due to GA₃ might have resulted in more production of photosynthates and their translocation through phloem to the root zone, which might be responsible for improving the root growth. Similar findings were reported by Wagh *et al.* (1998)^[26].

3.10 Fresh and dry weight of shoot and roots (g)

The maximum fresh and dry weight (4.13 g and 0.99 g) of shoots and roots (0.99 g and 0.49 g) respectively were obtained at 150 days after sowing under treatment M₅ comprising soil + vermicompost + AZO + PSB. Probable reasons may be that the media M₅ contains rich source of nutrient. The excellent plant growth in vermicompost was possibly due to some plant growth promoters in worm casts which increase physiological activities, essential for cell division, cell enlargement or both. The findings are supported by Roy and Shrivastava (2011)^[18] and Joshi *et al.* (2015)^[7]. Gibberellic acid had shown significant effect on fresh and dry

weight accumulation of shoot and root. Maximum fresh and dry weight of shoots (3.90 g and 0.93 g) and roots (0.92 g and 0.44 g) observed under 400 ppm concentration (G₂) at 150 DAS. Increase in fresh weight of roots is due to the influence of GA₃ on different plant parts, which could be due to its effect in stimulating cell division, cell elongation, auxin metabolism, cell wall plasticity and permeability of cell membrane leading to enhanced growth. Increase in the dry weight of different plant parts due to improved mobilization of nutrients due to the application of GA₃, which promotes plant growth and development. The findings are agreement with the findings of Rahemi and Baninasab (2000)^[15].

Table 5: Effect of growing media and gibberellic acid on root length, No of roots/seedling and Fresh and Dry weight of shoot and root at 150 DAS

Treatment	Root length (cm) at 150 DAS	No of roots/seedling at 150 DAS	Fresh weight (g) of shoot at 150 DAS	Dry weight (g) of shoot at 150 DAS (g)	Fresh weight (g) of root at 150 DAS	Dry weight (g) of root at 150 DAS
Gibberellic acid						
G ₀ (0 ppm)	21.44	45.13	3.26	0.73	0.74	0.38
G ₁ (200 ppm)	22.56	46.12	3.54	0.80	0.80	0.40
G ₂ (400 ppm)	24.52	48.90	3.90	0.93	0.92	0.44
G ₃ (600 ppm)	23.27	48.36	3.68	0.83	0.82	0.42
SEm	0.433	0.707	0.104	0.024	0.022	0.014
CD at 5% level	1.237	2.020	0.297	0.067	0.063	0.040
Media						
M ₀ (Soil)	18.76	41.45	2.88	0.63	0.67	0.29
M ₁ (Soil+FYM)	23.10	47.45	3.64	0.83	0.80	0.41
M ₂ (Soil+VC)	25.22	49.70	3.75	0.87	0.90	0.47
M ₃ (Soil+BF)	20.88	44.88	3.51	0.76	0.73	0.37
M ₄ (Soil+FYM+BF)	24.24	48.85	3.68	0.85	0.83	0.43
M ₅ (Soil+VC+BF)	26.13	50.37	4.13	0.99	0.99	0.49
SEm	0.530	0.866	0.127	0.029	0.027	0.017
CD at 5% level	1.574	2.474	0.364	0.082	0.077	0.049
Interaction						
G ₀ M ₀	17.35	39.47	2.34	0.58	0.64	0.24
G ₀ M ₁	21.56	46.20	3.34	.67	0.71	0.40
G ₀ M ₂	24.43	47.27	3.47	0.78	0.76	0.44
G ₀ M ₃	18.61	43.67	3.52	0.75	0.71	0.33
G ₀ M ₄	22.44	46.53	3.36	0.72	0.75	0.40
G ₀ M ₅	24.27	47.67	3.52	0.85	0.89	0.47
G ₁ M ₀	18.37	41.33	2.82	0.61	0.65	0.28
G ₁ M ₁	22.65	46.67	3.58	0.76	0.78	0.39
G ₁ M ₂	24.40	48.47	3.82	0.87	0.84	0.45
G ₁ M ₃	20.52	43.60	3.55	0.77	0.77	0.35
G ₁ M ₄	24.20	47.80	3.57	0.79	0.83	0.43
G ₁ M ₅	25.32	48.87	3.92	0.97	0.95	0.48
G ₂ M ₀	20.62	42.80	3.12	0.64	0.72	0.34
G ₂ M ₁	24.52	47.60	4.13	1.07	0.90	0.43
G ₂ M ₂	26.66	52.40	4.02	0.96	1.10	0.51
G ₂ M ₃	21.38	45.60	3.59	0.78	0.75	0.39
G ₂ M ₄	25.69	51.40	3.94	0.98	0.92	0.46
G ₂ M ₅	28.27	53.60	4.61	1.13	1.15	0.52
G ₃ M ₀	18.72	42.20	3.24	0.67	0.68	0.30
G ₃ M ₁	23.66	49.60	3.52	0.80	0.81	0.41
G ₃ M ₂	25.31	50.67	3.68	0.85	0.92	0.49
G ₃ M ₃	20.60	46.67	3.39	0.74	0.71	0.40
G ₃ M ₄	24.64	49.67	3.83	0.85	0.82	0.42
G ₃ M ₅	26.64	51.33	4.45	0.99	0.98	0.50
SEm	1.733	1.733	0.255	0.058	0.054	0.034
CD at 5% level	NS	NS	NS	NS	NS	NS

3. Conclusion

It is concluded that the growing media M₅ (soil + vermicompost + AZO + PSB) found superior over rest of the media, which significantly increased the germination and growth parameters. It is concluded that G₂M₅ combination [seed soaked in 400 ppm GA and sown in soil + vermicompost + AZO + PSB] was the best combination with respect to growth and survival of custard seedling and in

germination point of view, G₃M₅ treatment combination [seed soaked in 600 ppm GA concentration and sown in soil + vermicompost + AZO + PSB] was the best.

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