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The Pharma Innovation



ISSN (E): 2277-7695 ISSN (P): 2349-8242 NAAS Rating: 5.23 TPI 2023; 12(10): 1026-1030 © 2023 TPI

www.thepharmajournal.com Received: 18-07-2023 Accepted: 23-08-2023

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Effect of rooting substrates, PGR and biofertilizers on root characteristic, success and survivability of pomegranate (*Punica granatum* L.) cuttings cv. Bhagwa

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Abstract

An experiment entitled "Effect of Rooting Substrates, PGR and Biofertilizers on root characteristic and success &survivability of Pomegranate (*Punica granatum* L.) Cuttings cv. Bhagwa" was carried out during 2020-21 at RVSKVV – College of Horticulture, Mandsaur (MP). Experiment were consisting seventeen treatments and these treatments were replicated three times in CRD (Completely Randomized Design) and analyzed. Data raveled that treatment RS₁₇ (Soil + Vermicompost + Poultry Manure + IBA @ 2500 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g) show the best performance with respect of all of the parameters *i.e.* rooting percentage, length of longest root per cutting, diameter of thickest root per cutting, number of primary roots and secondary roots per cutting, fresh weight and dry weight of roots per cutting, success percentage of hardwood cutting and survival percent of hardwood cutting. RS1 (soil) (without IBA and Bio-fertilizers treatment) observed the lowest values of the traits likeroot characteristic and success & survivability.

Keywords: Pomegranate, root parameters, success percentage, survival percent

Introduction

Pomegranate (*Punica granatum* L.) is a well-known table fruit and it is cultivated in tropical and sub-tropical parts of the world. Pomegranate belongs to the family Punicaceae. Pomegranate is basically diploid in nature with 2n=2x=16 chromosomes (Jadhav and Sharma, 2007)^[12]. India is the world's largest producer of pomegranate. In India, occupies a cultivated area of 275 thousand hectares with 2,356 MT of production. In India pomegranate is mostly grown in Maharashtra (136.75 thousand ha.) (Anonymous, 2020)^[2].

Softwood cuttings, Air layering, Tissue culture, dormant hardwood cuttings and Grafting are all methods for vegetative propagation of pomegranates. Commercial propagation, on the other hand, is frequently done with hardwood cuttings of stem (Karimi, 2011; Polat and Caliskan, 2006) ^[13, 18]. Rooting in the vegetative propagation method like cutting is a major constraint. Application of plant growth regulators may be an effective technique for increasing the rooting ability and the growth of vegetative fragments cuttings.

Among phytohormones, auxin plays an essential role in regulating root development. The application of Indole Butyric Acid (IBA) induce rooting in stem cuttings due to their ability to activate cambium regeneration, cell division and cell multiplication (Rymbai and Reddy, 2010)^[24]. Besides this, rooting media is one of the most important factors for better rooting of cutting and survival of plant. There are different type of media like soil, FYM, vermi-compost, poultry manure, sand, perlite, vermiculite, *etc.* (Rathwa *et al.* 2017)^[22].

Vermicompost, which is high in organic manure due to the presence of bioactive principles. Poultry manure (chicken manure), is an excellent soil amendment that provides nutrients for growing crops and also improves soil quality (Ali, 2005)^[1]. The combined effect of biofertilizers and growth regulator have showed better root and shoot parameters which could be attributed to increased levels of growth promoting substance available due to synergistic effect of both biofertilizer and growth regulators in various ways (Damar *et al.* 2014)^[5]. *Trichoderma viride* and *Pseudomonas* also found to induce roots in pomegranate by suppressing the attacks of several disease (Shah and Afiya, 2019)^[26].

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Micro-organism which helps to solubilize insoluble phosphate are called as phosphate solubilizing microorganisms (PSMs). PSB not only provide P for growth of plant it also helps in increasing efficiency of N fixation and accelerate the accessibility of other trace elements (Swaminathan *et al.* 2020)^[28].

Materials and Methods

The present investigation was carried out from February 2021 to April 2021 at the experimental field of Department of Fruit Science, College of Horticulture, Mandsaur (M.P.). The type of cuttings used was hard wood cuttings of uniform size (15-20 cm long) with 5-6 functional buds were taken from vigorous, healthy plants of pomegranate variety Bhagwa. The partially matured branches, 0.75-1.00 cm in thickness were taken for cutting preparation. Shortly after their preparation, cuttings were kept in water to maintain the amount of moisture until planting time. Then, the basal portion of the

cuttings was treated for 5 minutes with growth regulator (quick deep method). Whereas, Bio-fertilizers and Bio control agents @ 2.5 g were mixed as per treatment in the rooting medium. Two third parts of the treated cuttings were placed in the rooting media at a slight angle (about 60°) vertical to the plane.

Results and Discussion

Statistically analyzed data embodied in Table- 1, 2 & 3. The rooting substrates, plant growth regulators, PSB and *Trichoderma viride* significantly affected the root characters *i.e.* rooting percentage, length of longest root per cutting (cm), diameter of thickest root per cutting (mm), number of primary roots per cutting, number of secondary roots per cutting, fresh weight of roots per cutting (g) and dry weight of roots per cutting (g), success percentage and survival percentof pomegranate (*Punica granatum* L.) cuttings cv. Bhagwa.

Table 1: Effect of rooting substrates, PGR and biofertilizers on root characteristic of pomegranate (*Punica granatum* L.) cuttings cv. Bhagwa.

S. No.	Treatments	Rooting percentage	Length of longest root per cutting (cm)	Diameter of thickest root per cutting (mm)
RS ₁	Soil	40.27	6.03	0.73
RS ₂	Soil + V.C.	46.79	7.48	0.92
RS ₃	Soil + P.M.	45.90	7.24	0.80
RS ₄	Soil + IBA @ 2000 ppm	50.45	9.67	1.08
RS ₅	Soil + IBA @ 2500 ppm	52.40	10.56	1.24
RS ₆	Soil + PSB @ 2.5 g	47.35	7.79	0.96
RS ₇	Soil + <i>T. viride</i> @ 2.5 g	48.67	8.50	0.98
RS ₈	Soil + V.C.+ P.M.+ IBA @ 2000 ppm	54.47	11.14	1.44
RS9	Soil + V.C.+ P.M.+ IBA @ 2500 ppm	55.92	11.57	1.50
RS_{10}	Soil + V.C.+ P.M.+ PSB @ 2.5 g	51.29	10.17	1.17
RS11	Soil + V.C.+ P.M.+ <i>T. viride</i> @ 2.5 g	53.00	10.79	1.30
RS ₁₂	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + PSB @ 2.5 g	57.47	11.39	1.52
RS ₁₃	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + PSB @ 2.5 g	60.23	12.50	1.67
RS ₁₄	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + T. viride @ 2.5 g	59.69	12.25	1.58
RS ₁₅	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + T. viride @ 2.5 g	62.19	12.84	1.78
RS16	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + T. viride @ 2.5 g + PSB @ 2.5 g	63.53	13.09	1.86
RS 17	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + <i>T. viride</i> @ 2.5 g+ PSB @ 2.5 g	65.95	14.29	1.90
	S.Em. (±)	0.81	0.30	0.03
	CD (5%)	2.33	0.86	0.09

Table 2: Effect of rooting substrates, PGR and biofertilizers on root characteristic of pomegranate (Punica granatum L.) cuttings cv. Bhagwa.

S.No.	Treatments	No. of primary roots per cutting	No. of secondary roots per cutting	Fresh weight of roots per cutting (g)	Dry weight of roots per cutting (g)
RS_1	Soil	6.24	34.11	0.62	0.09
RS_2	Soil + V.C.	8.90	40.80	0.86	0.14
RS ₃	Soil + P.M.	8.12	39.97	0.79	0.11
RS ₄	Soil + IBA @ 2000 ppm	10.71	44.48	1.04	0.23
RS ₅	Soil + IBA @ 2500 ppm	12.00	47.30	1.21	0.47
RS ₆	Soil + PSB @ 2.5 g	10.47	40.77	0.93	0.17
RS ₇	Soil + T. viride @ 2.5 g	10.02	41.34	0.98	0.20
RS ₈	Soil + V.C.+ P.M.+ IBA @ 2000 ppm	12.62	48.67	1.33	0.60
RS ₉	Soil + V.C.+ P.M.+ IBA @ 2500 ppm	12.61	48.89	1.36	0.72
RS_{10}	Soil + V.C.+ P.M.+ PSB @ 2.5 g	11.99	44.56	1.13	0.32
RS_{11}	Soil + V.C.+ P.M.+ <i>T. viride</i> @ 2.5 g	12.41	44.24	1.29	0.48
RS_{12}	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + PSB @ 2.5 g	13.00	50.65	1.40	0.75
RS_{13}	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + PSB @ 2.5 g	13.67	51.23	1.51	0.86
RS_{14}	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + T. viride @ 2.5 g	13.34	50.72	1.46	0.82
RS_{15}	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + T. viride @ 2.5 g	13.82	52.04	1.58	0.91
RS_{16}	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + T. viride @ 2.5 g + PSB @ 2.5 g	14.01	52.65	1.61	0.94
RS 17	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + T. viride @ 2.5 g+ PSB @ 2.5 g	14.98	53.90	1.70	0.99
	S.Em. (±)	0.33	0.42	0.03	0.01
	CD (5%)	0.95	1.22	0.08	0.04

S.No.	Treatments	Success percentage of shoot cutting	Survival percent
RS ₁	Soil	45.10	36.13
RS ₂	Soil + V.C.	52.54	41.11
RS ₃	Soil + P.M.	49.42	10.02
RS ₄	Soil + IBA @ 2000 ppm	55.61	46.27
RS ₅	Soil + IBA @ 2500 ppm	57.72	48.25
RS ₆	Soil + PSB @ 2.5 g	52.00	43.19
RS ₇	Soil + <i>T. viride</i> @ 2.5 g	53.39	43.22
RS ₈	Soil + V.C.+ P.M.+ IBA @ 2000 ppm	59.47	50.42
RS ₉	Soil + V.C.+ P.M.+ IBA @ 2500 ppm	60.86	51.30
RS_{10}	Soil + V.C.+ P.M.+ PSB @ 2.5 g	56.50	47.87
RS ₁₁	Soil + V.C.+ P.M.+ <i>T. viride</i> @ 2.5 g	58.00	49.11
RS ₁₂	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + PSB @ 2.5 g	61.85	53.08
RS ₁₃	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + PSB @ 2.5 g	64.84	56.22
RS ₁₄	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + T. viride @ 2.5 g	63.01	55.03
RS 15	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + T. viride @ 2.5 g	65.80	57.73
RS ₁₆	Soil + V.C.+ P.M.+ IBA @ 2000 ppm + T. viride @ 2.5 g + PSB @ 2.5 g	66.79	59.08
RS 17	Soil + V.C.+ P.M.+ IBA @ 2500 ppm + T. viride @ 2.5 g+ PSB @ 2.5 g	70.36	61.22
	S.Em. (±)	1.18	1.04
	CD (5%)	3 39	3.00

 Table 3: Effect of rooting substrates, PGR and biofertilizers on success percentage and survival percentof pomegranate (*Punica granatum* L.) cuttings cv. Bhagwa.

Note: Where, V.C. = Vermi-compost; P.M. = Poultry Manure; *T. viride* = *Trichoderma viride*; PSB = Phosphate solubilizing bacteria

Rooting percentage

The highest rooting percent (65.95%) were recorded in treatment RS₁₇ (soil + vermicompost + poultry manure + IBA @ 2500 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g). However, the lowest rooting percentage (40.27%) was recorded in treatment RS₁ (soil). This may be due to biofertilizers increased level of growth promoting substance, available N₂, P₂O₅ and other nutrients with the application of *Azotobacter* and PSB due to synergistic effect of biofertilizers in various ways and increase the concentration of IBA, which increased the level of auxins resulted in earlier completion of physiological processes in rooting and sprouting of cuttings (Tanwar *et al.* 2020) ^[29]. These finding are found similar with the result of Barde *et al.* (2010) ^[4] in pomegranate and Dhua *et al.* (1980) ^[6] in jackfruit.

Length of longest root per cutting (cm)

The longest root length (14.29 cm) was recorded in treatment RS₁₇ (soil + vermicompost + poultry manure + IBA @ 2500 ppm + Trichoderma viride @ 2.5 g + PSB @ 2.5 g). While, the shortest root length (6.03 cm) was recorded in RS_1 (soil).The reason for recording longest root may be attributed to the action of auxin activity which might have caused hydrolysis and translocation of carbohydrates and nitrogenous substances towards the base of cuttings and resulted in accelerated cell division and cell elongation under suitable environment. Another possible reason may be due to the early formation of roots and more utilization of reserved food materials of the treated cuttings (Ghatnatti, 1997)^[8]. Above result are in conformity with Rathore et al. (2020) [21], Pawar et al. (2020) ^[17], Rajamanikam and Balamohan (2019) ^[20], Manila et al. (2017)^[14] and Damar et al. (2014)^[5] in pomegranate cuttings.

Diameter of thickest root per cutting (mm)

The treatment RS₁₇ (Soil + Vermicompost + Poultry Manure + IBA @ 2500 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g) recorded the thickest root diameter 1.90 mm, While treatments Rs₁₆ (Soil + Vermicompost + Poultry Manure +

IBA @ 2000 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g) 1.86 mm was *at par* with the treatment RS₁₇. The result obtained may be due to higher accumulation of photosynthesis metabolites and nutrients under the IBA treatment (Dhua *et al*, 1980) ^[6]. Plant pathogen suppressing microorganisms (*eg. Trichoderma viride, Pseudomonas flurescence*) are ecofriendly and secretes growth promoting hormones and secondary metabolites which help in mobilizing various micronutrients (Arshad *et. al*, 1998)^[3] which helps the plants to grow a healthy root system with an increased diameter of the roots. The results obtained are in accordance with the previous results of Rathore *et al.* (2020) ^[21], Patil *et al.* (2001) ^[16] in pomegranate cuttings and Rawat *et al.* (2004) ^[23] in grapes.

Number of primary roots per cutting

The maximum number of primary roots (14.98) were recorded with the treatment of RS₁₇ (soil + vermicompost + poultry manure + IBA @ 2500 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g). Whereas, the minimum number of primary roots per cutting (6.24) were recorded in RS₁ (soil). It may be due to the action of auxin which might have caused hydrolysis and translocation of carbohydrates and nitrogenous substances at the base of cuttings and resulted in accelerated cell elongation and cell division in suitable environment (Hartmann *et al.* 2002) ^[11]. These results are similar line with the finding of Mehta *et al.* (2018) ^[15] in pomegranate cuttings.

Number of secondary roots per cutting

The treatment RS₁₇ (Soil + Vermicompost + Poultry Manure + IBA @ 2500 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g) recorded the maximum mean number of secondary roots per cutting (53.90). It may be due to the action of auxin which might have caused hydrolysis and translocation of carbohydrates and nitrogenous substances at the base of cuttings and resulted in accelerated cell elongation and cell division in suitable environment (Hartmann *et. al,* 2002) ^[11]. These results are similar line with the finding of Mehta *et al.* (2018) ^[15] in pomegranate cuttings.

Fresh weight of roots per cutting (g): The maximum fresh weight of roots (1.70 g) was recorded with the treatment of RS₁₇ (soil + vermicompost + poultry manure + IBA @ 2500 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g). The feasible reason for increase root weight may be due to the production of more number of roots, increased length and diameter of roots which results in production of heavier roots which in turn results in increased root weight. The result obtained is in harmony with the results of Rathore *et al.* (2020) ^[21], Hakim *et al.* (2018) ^[10] in pomegranate cuttings and Sarita *et al.* (2019) ^[28] in layering of guava.

Dry weight of roots per cutting (g)

The treatment RS₁₇ (Soil + Vermicompost + Poultry Manure + IBA @ 2500 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g) recorded the maximum dry weight of roots per cutting (0.99 g). The feasible reason for increase root weight may be due to the production of more number of roots, increased length and diameter of roots which results in production of heavier roots which in turn results in increased root weight. The result obtained is in harmony with the results of Rathore *et al.* (2020) ^[21], Hakim *et al.* (2018) ^[10] in pomegranate cuttings and Sarita *et al.* (2019) ^[28] in layering of guava.

Success percentage of shoot cutting

Significantly maximum success percentage (70.36%) of shoot cutting were recorded in RS₁₇ (soil + vermicompost + poultry manure + IBA @ 2500 ppm + Trichoderma viride @ 2.5 g + PSB @ 2.5 g). However, minimum success percentage of shoot cutting (45.10%) was recorded under treatment RS_1 (soil). Success percentage of cuttings depends on several factors like formation of adventitious roots, developments of healthy root system, presence of leaves, type of rooting media, presence of auxin etc. (Gautherest, 1969) [7]. The higher success percentage might be due to higher accumulation of photosynthetic metabolites and nutrients. The more and healthy root formation by the exogenous application of IBA with media one of the possible reason for enhancing the success percent of cuttings. Biofertilizers have a tendency to increase the level of nutrients, plant growth regulators and vitamins for the plants (Slankis, 1973)^[27]. The results are in agreement with the earlier findings of Ramanickam and Balamohan (2019)^[20], Gurjar and Patel (2007)^[9], Upadhyay and Badyal (2007)^[30] in stem cutting of pomegranate.

Survival Percent

Significantly maximum survival percent of shoot cutting (61.22%) was recorded in treatment RS_{17} (Soil + Vermicompost + Poultry Manure + IBA @ 2500 ppm + Trichoderma viride @ 2.5 g + PSB @ 2.5 g), while treatment Rs₁₆ (Soil + Vermicompost + Poultry Manure + IBA @ 2000 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g) 59.08%, was at par with the treatment RS_{17} . The survival of the sprouted cuttings might be directly linked to the formation of adventitious roots on cuttings. The result may be due to high carbohydrate reserves per cutting and optimum concentration of IBA. The same factors brought about maximum number of shoots and roots per cutting and root length which in turn contributed to high survival percentage (Purohit and Shekharappa, 1985)^[19]. Trichoderma and Pseudomonas being biological agents provide resistance to biotic and abiotic stress and increase the root growth, uptake and use of nutrients to the plants and increases the percentage of several major root

fungi like pythium. *Trichoderma* kills *Rizoctonia and Fusarium* and results in a healthy root development by decreasing the attack of soil borne pathogens, which in turn results in increased survival of the cuttings. The results are in agreement with the earlier findings of Rathore *et al.* (2020) ^[21], Gurjar and Patel (2007) ^[9] in stem cutting of pomegranate.

Conclusion

The treatment of RS_{17} (Soil + Vermi-compost + Poultry Manure + IBA @ 2500 ppm + *Trichoderma viride* @ 2.5 g + PSB @ 2.5 g) was found to be the most efficacious in invigorating the root parameters (*i.e.* rooting percentage, length of longest root per cutting, diameter of thickest root per cutting, number of primary roots and secondary roots per cutting, fresh weight and dry weight of roots per cutting), success percentage of cutting and survival percent of hardwood cutting in pomegranate cv. Bhagwa.

Acknowledgement

With profound respect I am extremely thankful to Dr. S.K. Rao, Hon'ble Vice Chancellor, R.V.S.K.V.V., Gwalior, Dr. A.K. Singh, Director of Instruction and Dr. D.H. Ranade, Dean Faculty of Agriculture R.V.S.K.V.V., Gwalior, (M.P.) with deepest sense of humility and gratefulness, I feel myself duly bound to express my heartfelt and sincere thanks to Dr. Mridula Billore, Dean, KNK College of Horticulture, Mandsaur. The words at my dominion are really scarce to articulate my deep sense of appreciation to Dr. R.N. Kanpure, Associate Professor, Department of Fruit Science.

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