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Effect of biofertilizers and CSR-BIO on the growth of clonal Dogridge (*Vitis champini*) rootstock

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Abstract
An experiment was carried out to elucidate the effect of biofertilizers (Azospirillum and Phosphobacteria) and CSR-BIO alone and their combination on growth parameters of dogridge (*Vitis X champini)* rootstock at Grapes Research Station, Anaimalayanpatty, Tamil Nadu, India. The experiment was conducted in completely randomized design (CRD) with 16 treatments (T_1 (Azospirillum 1 g / polybag), T_2 (Azospirillum 2 g/ polybag), T_3 (Phosphobacteria 1 g/ polybag), T_4 (Phosphobacteria 2 g / polybag), T_5 (CSR- BIO 1 g / polybag), T_6 (CSR- BIO 2 g / polybag), T_7 (T_1 + T_3+ T_5), T_8 (T_1 + T_3+ T_6), T_9 (T_1 + T_2+ T_5), T_{10} (T_1 + T_4+ T_6), T_{11} (T_2 + T_3+ T_5), T_{12} (T_2 + T_3+ T_6), T_{13} (T_2 + T_4+ T_5), T_{14} (T_2 + T_4+ T_6), T_{15} (with IBA dipping) and T_{16} (without IBA dipping)) and three replications. The data revealed that T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR- BIO 2 g/ polybag) showed the highest values for sprouting per cent (96.67), survival per cent (95.00), stem diameter (2.35 mm), fresh weight of shoot (5.09 g), fresh weight of root (1.82 g), number of primary roots (6.50) and shoot/root ratio (2.80). The same treatment T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR- BIO 2 g/ polybag) also expressed the highest number of leaves cutting^{-1} (18.13), shoot length (27.56 cm) and root length (35.15 cm). While control (T_{16}) recorded the lowest values for all growth parameters.

Keywords: Biofertilizers, CSR-BIO microbial culture, dogridge, propagation, rootstock

1. Introduction
The grapes (*Vitis vinifera* L.) is one of the most important fruit crop of tropical and subtropical regions in the world. It belongs to the family Vitaceae. It is believed to have been originated in Armenia near the Caspian Sea and got introduced into India via Iran and Afghanistan during the 11th century. Though this was naturally originated as a temperate plant, subsequently acclimatized to tropical and sub-tropical conditions of Peninsular India (Shikhamany 2001). The total area is 0.13 million hectares with an annual production of 2.95 million tonnes (NHB 2019). Grape is commercially propagated through stem cuttings. The cuttings are the simplest and most extensively used form of asexual propagation. Exogenous growth hormones have been proven to be the most effective in promoting early and better root development in cuttings. Hardwood cuttings are made from shoots that are one season old and have three to four buds. Dogridge is one of the commercially important rootstocks used in commercial grapes cultivation because of its drought and salinity tolerance. Moreover, it can also exhibit tolerance against nematodes and helps in better nutrient uptake from the soil on grafted vines. Multiplication of dogridge rootstock through hardwood stem cutting is the easiest method to get uniform plants in a shorter period. In general, the rooting ability of stem cuttings depending upon species, location, season, biochemical composition of the mother wood and age (Satisha *et al.*, 2007) [11]. Some of the internal factors *viz.*, age, tissue maturity, bud position, number of leaves and the amount of stored food in cuttings are determinant for formation of callus tissue and adventitious roots initiation which affect the rooting capacity of cuttings. Further, rooting media is also one of the most important factors affecting rooting and growth of grapes cuttings. The success rate of dogridge cuttings depends on many factors *viz.*, mother plant character, internal factor related to mother plant, maturity of wood, season, temperature and relative humidity. It has been very well demonstrated that the grapes cutting would take longer period for sprouting is poor. Auxin is used for root development in hardwood stem cutting by accelerating the formation of adventitious roots which positively enhanced the survival percentage (Galavi *et al.*, 2013) [7]. *Azospirillum*, is a gram-negative free-living nitrogen-fixing rhizobacteria, which extensively used for nitrogen fixation and phytohormone production, especially auxin-related compounds *viz.*, indole-3-acetic acid (IAA) (Dobbelaere *et al.*, 2001; Bashan *et al.*, 2004) [3].

Application of PGPR in vegetatively propagated plants improved the root development along with a better root surface area and root biomass production (Carletti, 2002). The use of auxins at higher concentration initiates the early development of roots and, therefore, increased nutrients uptake due to increased root length. The CSR-BIO microbial culture acts as a nutrient mobilizer, growth enhancer, soil vitalizer and plant protectant for crops under normal and alkaline soil condition.

The CSR-BIO microbial consortium was developed from rhizosphere microbial diversity at Central Soil Salinity Research Institute, Lucknow. The beneficial effect of CSR-BIO has been very well studied and documented in various horticultural crops for improving the yield and quality (Damodaran *et al.*, 2013) [5]. With this background, the present study was carried out to elucidate the effect of CSR-BIO microbial culture and biofertilizers on growth and development of clonal dogridge (*Vitis X champini*) rootstock.

2. Materials and Methods

2.1. Plant material and Experimental site

The present experiment was conducted to study the rooting behaviour of clonal dogridge (*Vitis champinii*) rootstock at Grapes Research Station (TNAU), Anaimalayanpatty, Rayappanpatty, Theni district, Tamil Nadu, India during the period 2019-2020. Hardwood cuttings of dogridge with three to four well developed buds were collected and uniform slant cut was given in the basal portion of the hardwood cuttings and dipped in a solution containing Indole 3-butyric acid @ 2000 ppm for 30 seconds except control.

2.2. Experimental details

The nursery experiment was laid out in complete randomized design (CRD) with 16 treatment combinations consisting of biofertilizers and CSR-BIO *viz.*, (T_1 (Azospirillum 1 g / polybag), T_2 (Azospirillum 2 g/ polybag), T_3 (Phosphobacteria 1 g/ polybag), T_4 (Phosphobacteria 2 g / polybag), T_5 (CSR-BIO 1 g / polybag), T_6 (CSR-BIO 2 g / polybag), T_7 (Azospirillum @ 1g + Phosphobacteria @ 1g + CSR-BIO @ 1g / polybag), T_8 (Azospirillum @ 1g + Phosphobacteria @ 1 g + CSR-BIO @ 2 g / polybag), T_9 (Azospirillum @ 1 g + Phosphobacteria @ 2 g + CSR-BIO @ 1 g / polybag), T_{10} (Azospirillum @ 1 g + Phosphobacteria @ 2 g + CSR-BIO @ 2 g / polybag), T_{11} (Azospirillum @ 2 g + Phosphobacteria @ 1 g + CSR-BIO @ 1 g / polybag), T_{12} (Azospirillum @ 2 g + Phosphobacteria @ 1 g + CSR-BIO @ 2 g / polybag), T_{13} (Azospirillum @ 2 g + Phosphobacteria @ 2 g + CSR-BIO @ 1 g / polybag), T_{14} (Azospirillum @ 2 g + Phosphobacteria @ 2 g + CSR-BIO @ 2 g / polybag), T_{15} (IBA @ 2000 ppm dipping) and T_{16} (control)) in three replications. One hundred cuttings were taken for each treatment in each replication in polybags of size 5”X7” filled with a rooting media consist of red soil, farmyard manure and sand (1:1:1) in equal proportion and kept under shade net house.

2.3. Growth parameters

Observations on growth parameters *viz.*, sprouting per cent, survival per cent, number of leaves cutting^{-1}, stem diameter (mm), shoot length (cm), fresh weight of shoot (g), fresh weight of root (g), number of primary roots, shoot/root ratio and root length (cm) were recorded at 90 days after planting. The data were subjected to statistical scrutiny by using standard ANOVA.

3. Results

The growth parameters *viz.*, sprouting per cent, survival per cent, number of leaves cutting^{-1}, stem diameter (mm), shoot length (cm), fresh weight of shoot (g), fresh weight of root (g), number of primary roots, shoot/root ratio and root length (cm) were recorded and presented in table 1 and 2. The statistical analysis of growth parameters from different treatments showed significant variations. The success percent findings revealed a considerable difference between the various treatments. The current investigation findings revealed that hardwood cuttings treated with Azospirillum 2 g + Phosphobacteria 2 g + CSR-BIO 2 g/ polybag (T_{14}) had the most effective sprouting per cent of 96.67%, significantly higher than all other treatments. This might be owing to the IBA treatment, which significantly reduced the time it took for cuttings to sprout. The findings of the study revealed that Azospirillum 2 g + Phosphobacteria 2 g + CSR-BIO 2 g/ polybag (T_{14}) exhibited the maximum survival per cent (95.00).

The results showed the highest values for growth parameters *viz.*, number of shoots cutting^{-1} (1.78), stem diameter (2.35 mm), fresh weight of shoot (5.09 g), fresh weight of root (1.82 g), number of primary roots (6.50) and shoot/root ratio (2.80) were registered in the treatment T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR-BIO 2 g/ polybag). The same treatment T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR-BIO 2 g/ polybag) also recorded the highest number of leaves (18.13), shoot length (27.56 cm) and root length (35.15 cm). The results revealed that the biofertilizers and CSR-BIO microbial culture showed non-significant variation on number of shoots cuttings^{-1}. While the treatment T_{16} (control) showed the lowest values for yield parameters *viz.*, sprouting per cent (73.30), survival per cent (66.26), stem diameter (1.78 mm), fresh weight of shoot (2.03 g), fresh weight of root (0.91 g), number of primary roots (2.00), shoot/root ratio (2.50) and root length (11.20 cm).

4. Discussion

The data illustrated that T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR- BIO 2 g/ polybag) had the highest sprouting per cent and survival per cent and it was significantly different from all other treatments. The highest sprouting per cent and survival per cent might be due to increased availability of endogenous plant growth regulators especially auxin like compounds in the cutting which in turn helps in early completion of physiological processes *i.e.*, rooting and sprouting of cutting. This fact has been supported by the application of biofertilizers which increased the plant growth regulators levels in plants and success rate (Slankis, 1973) [15]. African Blackwood basal cuttings treated with IBA sprouted earlier than untreated cuttings, according to Amri *et al.* (2010) [1]. These findings were corroborated with the earlier works of Barde *et al.* (2010) [2] and Sharma and Bhutani (1998) [12]. T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR- BIO 2 g/ polybag) exhibited the highest number of shoots cuttings^{-1}, whereas the lowest number of shoots cuttings^{-1} was found in T_{16} (Control). This might be due to the positive effect of plant growth promotors on the number of shoots and vigorous root system. An increase in number of shoots per cutting and number of leaves per shoots by 1500 ppm IBA in sweet lime (*Citrus limettaides*) was reported by Singh and Pande (1986). The highest number of primary roots cutting^{-1}, fresh root weight and length of roots were found in the treatment Azospirillum 2 g + Phosphobacteria 2 g + CSR-

BIO 2 g/ polybag (T_{14}). This might be due to increased synthesis of growth promoting substances and availability of more phosphorus which enhanced the rooting ability in cuttings. In cinnamon propagation, application of phosphobacteria at the time of planting recorded the maximum values for rooting per cent, number of roots cutting^{-1} (Nageswari *et al.*, 1999) [9]. Wange and Ranawade (1997) [16] found that the maximum rooting percentage was obtained in grapes cutting treated with biofertilizers. The longer root length in hardwood cuttings may be due to more significant starch buildup, which creates a favourable environment for root initiation. The findings are consistent with those of Shekharappa (1983) [13] in pomegranate, Reddy *et al.* (2008) in fig, and Amri *et al.* (2010) [1] in African Blackwood. Total number of leaves cutting^{-1}, shoot length and stem diameter were found significantly maximum in T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR- BIO 2 g/ polybag), whereas the lowest number of leaves cutting^{-1} was recorded in T_{16} (control). This might be due to higher photosynthetic activity of leaves, metabolites and nutrients supplement. The increased number of leaves cutting^{-1} might be due to vigorous root system which helps to increase the nutrients uptake. While the treatment T_{16} (control) showed the lowest values for sprouting per cent, survival per cent, number of leaves cutting^{-1} and root length. These poor results might be due to less growth substance with low physiological activity for triggering root initiation and development. Exogenous application of IBA @ 1500 ppm showed superior results in terms of root characteristics (Kumawat *et al.*, 2010) [8]. The similar trend was also reported by Kale and Bhujbal (1971) in which IBA @ 1500 ppm recorded the highest value for rooting of cuttings, primary roots, length of shoot and number of the leaves in Bougainvillea.

Table 1: Effect of biofertilizers and CSR-BIO microbial culture on growth parameters in dogridge rootstock (90 days after planting)

Treatments	Sprouting per cent	Survival per cent	Number of leaves cutting^{-1}	Number of shoots cuttings^{-1}	Stem diameter (mm)	Shoot length (cm)
T_1	83.92	70.37	10.80	1.33	2.04	17.21
T_2	87.33	82.30	14.80	1.57	1.98	20.05
T_3	84.40	71.17	10.20	1.32	2.00	17.99
T_4	86.53	76.61	16.40	1.50	1.92	21.48
T_5	87.07	76.40	11.00	1.45	1.88	17.44
T_6	88.89	81.74	14.60	1.41	1.94	22.19
T_7	80.22	73.77	16.73	1.44	1.86	19.19
T_8	85.00	73.33	16.53	1.47	2.09	23.63
T_9	91.00	73.98	13.00	1.42	1.96	20.67
T_{10}	91.67	65.03	11.93	1.58	2.12	25.44
T_{11}	77.22	82.00	13.93	1.52	2.01	22.25
T_{12}	95.60	92.63	18.13	1.74	2.12	27.56
T_{13}	93.16	86.31	14.93	1.41	2.09	21.99
T_{14}	96.67	95.00	16.87	1.78	2.35	27.28
T_{15}	75.43	69.67	10.27	1.33	2.05	17.01
T_{16}	73.30	66.26	9.60	1.30	1.78	16.15
CD (0.05)	11.72	9.19	3.74	NS	0.17	6.88
SE (d)	5.83	4.49	1.83	0.15	0.08	3.35

Table 2: Effect of biofertilizers and CSR-BIO microbial culture on growth parameters in dogridge rootstock (90 days after planting)

Treatments	Fresh weight of shoot (g)	Fresh weight of root (g)	Shoot/ root ratio	Number of primary roots cutting^{-1}	Root length (cm)
T_1	2.59	0.96	2.69	2.50	18.19
T_2	3.26	1.45	2.25	3.00	24.75
T_3	3.36	1.05	3.21	2.50	18.33
T_4	3.12	1.22	2.59	4.00	22.48
T_5	3.17	1.10	2.93	2.50	17.51
T_6	3.37	1.40	2.44	3.00	26.55
T_7	3.20	1.55	2.15	3.50	27.77
T_8	3.44	1.77	1.95	4.00	20.16
T_9	4.47	1.32	3.38	3.00	18.40
T_{10}	3.03	1.50	2.02	5.50	29.45
T_{11}	3.96	1.46	2.71	5.00	20.60
T_{12}	3.73	1.79	2.09	6.00	35.15
T_{13}	4.34	1.69	2.71	6.00	23.85
T_{14}	5.09	1.82	2.80	6.50	30.60
T_{15}	2.25	0.93	2.56	2.00	14.95
T_{16}	2.03	0.91	2.50	2.00	11.20
CD (0.05)	0.70	0.28	0.73	1.36	5.85
SE (d)	0.34	0.14	0.36	0.67	2.89

5. Conclusion

From the present investigation, it is highlighted that application of biofertilizers and CSR-BIO microbial culture enhanced the growth and rooting parameters of dogridge (*Vitis* X *champini*) rootstock. The data revealed that T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR- BIO 2 g/ polybag) showed the highest values for sprouting per cent, survival per cent, number of cutting^{-1}, stem diameter, fresh weight of shoot, fresh weight of root, number of primary roots and shoot/root ratio. The same treatment T_{14} (Azospirillum 2 g + Phosphobacteria 2 g + CSR- BIO 2 g/ polybag) expressed the highest number of leaves, shoot length and root length. While control (T_{16}) recorded the lowest values for all growth parameters.

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