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Biostimulants as Eco-friendly Inputs for Improving Yield, Fruit Quality and Orchard Soil Properties of Pomegranate (*Punica granatum* L.)

Sajjadullah[1], Naveen C. Sharma[1], Priyanka Chauhan[1], Gopal Singh[1] and Uday Sharma[2]

[1]Dept. of Fruit Science, College of Horticulture, [2]Dept. of Soil Science and Water Management, College of Forestry, Dr Y S Parmar University of Horticulture and Forestry, Nauni (173 230), India

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Corresponding naveenuhf@gmail.com

0000-0003-0910-5792

ABSTRACT

An experiment was carried out during December 2019–December 2020 to study the effect of different doses of commercial biostimulant formulations on yield, fruit quality and orchard soil properties of pomegranate. Eleven years old trees planted at 4.0×2.0 m^2 spacing were selected for the studies. The experiment was laid out in a randomized block design with 13 treatments comprising different doses of granulated biostimulant (100, 200 and 300 g $plant^{-1}$) as soil application and foliar sprays of liquid biostimulant (1.5 ml l^{-1}) alone or in combination. Results revealed that fruit yield, quality and soil properties were significantly influenced by different doses of biostimulant formulations. The maximum yield, fruit length, breadth, fruit weight, aril weight, total soluble solids, juice content, total sugars and lowest titratable acidity were recorded in trees treated with Biostimulant (B_1) @ 300 g $plant^{-1}$ as two soil applications of 150 g each+two foliar sprays of Biostimulant (B_2) @ 1.5 ml l^{-1} (T_{12}). However, maximum soil organic carbon, available N, P, K, bacterial, fungi, and actinomycetes, were observed significantly higher with Biostimulant (B_1) @ 300 g $plant^{-1}$ as single soil application+one foliar spray of Biostimulant (B_2) @ 1.5 ml l^{-1} (T_9) and closely followed by Biostimulant (B_1) @ 300 g $plant^{-1}$ as two soil applications of 150 g each+two foliar sprays of Biostimulant (B_2) @ 1.5 ml l^{-1} (T_{12}). Thus, biostimulant (B_1) @ 300 g $plant^{-1}$ as two soil applications of 150 g each+two foliar sprays of biostimulant (B_2) @ 1.5 ml $litre^{-1}$ (T_{12}) was found as most effective treatment for improving yield, fruit quality and orchard soil properties of pomegranate.

KEYWORDS: Biostimulant, fruit quality, microbial count, pomegranate, soil nutrients, yield

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1. INTRODUCTION

Biostimulants are preparations of biological origin which stimulate natural ecological processes to improve nutrient absorption and nutritional quality of plants (Yakhin et al., 2017). Biostimulants are known to improve fruit size, appearance and aril quality by direct effects on fruit growth and development or indirectly by regulating crop load, tree vigour and canopy architecture (Looney, 1993). Biostimulants and bioprotectants can also reduce the abiotic and biotic stresses impact and decrease the cropping systems' dependency on chemical fertilizers and pesticides (Bulgari et al., 2019; Cuadrado et al., 2019).

Biostimulant formulations containing sea weed extract, humic acid, ascorbic acid and amino acids are commercially available in the market and their use is relatively cheaper and economical to the farmers. Sea weed extract contains phytohormones and growth regulators like auxins, gibberellins, cytokinins, abscisic acid, ethylene, betaine, polyamines, vitamins, amino acids, antibiotics and micronutrients (Panda et al., 2012), which helps in increasing the plant growth, inducing resistance against pathogens and thriving under extreme weather conditions. Sea weed extract has been found responsible for causing a broad range of useful effects like increased yield, fruit quality, shelf life of the produce, nutrient uptake, resistance to frost stress conditions and reduced incidence of fungal and insect attack (Metting et al., 1990). Additionally, the application of sea weed extract has also been associated with an increase in plant tolerance to biotic and abiotic stresses (Holdt and Kraan, 2011; Guinan et al., 2012; Battacharya et al., 2015). Several studies in the recent times indicate a positive influence of sea weed extract in crop production in both normal and stress conditions (Carillo et al., 2020; Lee and Ryu, 2021; Pohl et al., 2019). Humic substances are the most active component of organic matter and soil, which is responsible for several complex chemical reactions in the soil (Trevisan et al., 2019). Humic acid stimulates soil microbial activity, transform insoluble nutrients into soluble form and mobilizes plant nutrients more effectively and making them available to the plants. Humic acid comprises of 42–46% oxygen, 44–58% carbon, 0.5–4.0% nitrogen and 6-8% hydrogen (Larcher, 2004) and induce morphological, physiological, genetic and biochemical effects (Chen et al., 2004). The beneficial effects of humic substances include lateral root development and root hair formation in fruit crops (Ramos et al., 2015). Humic acid has a role in drought stress which is one of the most impairing stresses affecting crop growth and productivity by reducing metabolic and enzymatic activity in plants (Farooq et al., 2009; Selmar et al., 2013; Hatfield and Dold, 2019). Under water deficit conditions, the addition of humic acid to the soil reduce the negative effects of stress by improving the production of photosynthetic pigments while keeping the relative water content at higher levels (Abdelaal et al., 2018; Khorasaninejad et al., 2018; Qin et al., 2018; Manhong et al., 2020).

Ascorbic acid is an essential ingredient for metabolic reactions in plants and help plants to overcome stress. It acts as a major redox buffer and cofactor for enzymes and involved in regulating photosynthesis, hormone biosynthesis, regenerating other antioxidants, cell division and improving the nutritional quality (Gallie, 2012) thus, considered essential for plants. It can serve as a source of carbon and energy, which protect the plants against pathogens. Biostimulants can act on soil nutrient availability and uptake through different ways that affect soil processes. This may include improvement of soil structure and increase of soil capacity to exchange cations, improvement of micronutrients by increasing their solubility and preventing their leakage, and adjustment of the soil pH (Rouphael and Colla, 2018). Keeping in view, the importance of biostimulants and the role they play in improving plant's growth and development, the present study was conducted to investigate the effect of eco-friendly biostimulant formulations on yield, fruit quality and orchard soil properties of pomegranate.

2. MATERIALS AND METHODS

The present investigation was conducted at Model Farm of Dr. Y S Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh, India during the year December 2019–December 2020. The experimental site was situated at an elevation of 1250 m above mean sea level with latitude of 30° 50' 45" N and longitude of 77° 88' 33" E. The annual rainfall of the area ranges between 800–1500 mm, major amount of which is received during monsoon period from July to September. Eleven-years-old healthy trees of pomegranate cv. Kandhari which were uniform in size and vigour and planted at a spacing of 4.0×2.0 m^2 apart were randomly selected for the experiment. Uniform cultural practices were given to each experimental tree during the course of study to keep the plants in good health. The experiment was laid out in a randomized block design with thirteen treatments viz. Biostimulant (B_1) @ 100 g $plant^{-1}$ as single soil application (T_1), Biostimulant (B_1) @ 200 g $plant^{-1}$ as single soil application (T_2), Biostimulant (B_1) @ 300 g $plant^{-1}$ as single soil application (T_3), Biostimulant (B_1) @ 100 g $plant^{-1}$ as two soil applications of 50 g each (T_4), Biostimulant (B_1) @ 200 g $plant^{-1}$ as two soil applications of 100 g each (T_5), Biostimulant (B_1) @ 300 g $plant^{-1}$ as two soil applications of 150 g each (T_6), T_1+one foliar spray of Biostimulant (B_2) @ 1.5 ml l^{-1} (T_7), T_2+one foliar spray of Biostimulant (B_2) @ 1.5 ml l^{-1} (T_8), T_3+one foliar spray of Biostimulant (B_2) @ 1.5 ml $litre^{-1}$ (T_9), T_4+two foliar sprays of Biostimulant (B_2) @ 1.5 ml $litre^{-1}$

(T_{10}), T_5+two foliar sprays of Biostimulant (B_2) @ 1.5 ml l^{-1} (B_{11}), T_6+two foliar sprays of Biostimulant (B_2) @ 1.5 ml l^{-1} (T_{12}) and Control (T_{13}). Each treatment was replicated three times. The commercial formulation of granular biostimulant (B_1) had humic acid (39%), kelp extract (25%), amino acids (9.0%), ascorbic acid+thiamine+inositol+to copherol (27%) as its constituents, whereas, commercial formulation of liquid biostimulant (B_2) contained sea weed extract. Granular biostimulant (B_1) was applied at the time of initiation of flowering (single application) and time of initiation of flowering as well as 45 days after 1st application (two applications) by broadcasting to the basin soil of respective trees as per treatments. However, liquid biostimulant (B_2) was applied as foliar spray at 15 days after initiation of flowering (single spray) and 15 days after initiation of flowering as well as 45 days after initiation of flowering (two sprays). Biostimulant was not applied to the control trees but foliar water sprays were given at the time of biostimulant sprays to the treatment trees. The biostimulant solution was sprayed at low volume with knapsack sprayer during morning hours on foliage, flowers and fruitlets covering the whole canopy of the plant.

All the fruits harvested at maturity from individual trees were weighed to calculate the yield per tree. Ten fully mature fruits were collected randomly from the periphery of each experimental plant at the time of harvest and brought to the laboratory for physico-chemical analysis. Length and diameter of randomly selected fruits was measured between calyx and stylar end of the fruit with the help of digital vernier calipers. The fruits under each treatment were weighed using an electronic top pan balance. Volume of the fruit was measured using water displacement method. The rind thickness of fruits from each sample was measured with the help of digital vernier calipers at four different positions after cutting the fruit breadth wise. The aril of the fruits under each replication was separated and weighed on a digital top pan balance. The total soluble solids of the fruit juice were determined with Erma-Hand refractometer (0 to 32°B). Titratable acidity was calculated with the help of formula given by (Ranganna, 1995). The sugar content of the fruit was determined as per method suggested by Ranganna (1995). The quantitative determination of ascorbic acid was done as per the method prescribed by AOAC (1980). Soil pH and EC was determined using method given by (Jackson, 1973). Organic carbon in the soil sample was estimated by wet digestion method of Walkley and Black (1934). Alkaline potassium permanganate method developed by Subbiah and Asija (1956) was used to determine available nitrogen content in soil. Phosphorus in soil was determined following procedure given by (Olsen et al., 1954). Available potassium content in the soil was extracted with neutral ammonium acetate as per the procedure given by Merwin and Peech (1951). Standard plate count technique was used for the total microbial count (Wollum, 1982).

3. RESULTS AND DISCUSSION

Biostimulant applications at different doses induced significant effects on yield and fruit quality characteristics of pomegranate (Table 1). Fruit yield and physical quality characteristics got improved with the increasing doses of biostimulants. All the biostimulant treatments significantly increased the fruit yield as compared to control. However, maximum yield (19.17 kg tree^{-1}) was recorded under T_{12} (Biostimulant B_1 @ 300 g plant^{-1} as two soil applications of 150 g each+two foliar sprays of Biostimulant B_2 @ 1.5 ml l^{-1}), closely followed by T_{11} (18.67 kg tree^{-1}). Fruits of biggest size (85.09 mm length×89.15 mm diameter) and maximum weight (413.33 g) as well as volume (430.00 cc) were obtained from the trees treated with T_{12} (Biostimulant B_1 @ 300 g plant^{-1} as two soil applications of 150 g each+two foliar sprays of Biostimulant B_2 @ 1.5 ml l^{-1}). This treatment was statistically at par with treatment T_{11} with respect to fruit length, whereas, it was found statistically at par with treatments T_9, T_{10} and T_{11} with respect to fruit diameter, weight and volume. Similarly, aril weight and juice content in the fruits increased with the increasing doses of biostimulants. The maximum aril weight (318.81 g) and juice content (74.00%) were recorded under T_{12} (Biostimulant B_1 @ 300 g plant^{-1} as two soil applications of 150 g each+two foliar sprays of Biostimulant B_2 @ 1.5 ml l^{-1}). Treatment T_{12} had significantly higher aril weight as compared to all the treatments of biostimulant B_1 as soil application (T_1 to T_6), combinations of biostimulant B_1 as soil application+single foliar spray of biostimulant B_2 (T_7 to T_9) and control (T_{13}). However, with respect to juice content T_{12} was significantly superior to all the treatments of biostimulant B_1 as single soil application (T_1 to T_3), biostimulant B_1 @ 100 and 200 g in two soil applications (T_4 to T_5) and control (T_{13}). Rind thickness of the fruit was not influenced by the application of biostimulants. Soil application of biostimulant B_1 in two split doses was found more effective as compare to single application to improve the yield and fruit physical quality characteristics, whereas, combination of soil application of biostimulant B_1+two foliar sprays of biostimulant B_2 was more effective as compared to combination of soil application of biostimulant B_1+one foliar spray of biostimulant B_2. The minimum yield, fruit size, fruit and aril weight, fruit volume and juice content were observed in fruits harvested from untreated (control) trees. The increase in fruit yield and improvement in physical fruit quality characteristics with the application of biostimulant doses might be due to the presence of humic acid in biostimulants, which increases N uptake and

Table 1: Effect of biostimulants on yield and fruit physical quality characteristics of pomegranate

Treatment	Fruit yield (kg tree^{-1})	Fruit length (mm)	Fruit diameter (mm)	Fruit weight (g)	Fruit volume (cc)	Aril weight (g)	Juice content (%)	Rind thickness (mm)
T_1	13.17	74.17	78.04	285.07	294.17	201.73	64.33	4.11
T_2	14.10	74.88	79.29	300.48	310.33	221.67	66.00	3.91
T_3	14.47	75.41	80.27	319.57	331.13	233.73	66.33	3.64
T_4	13.50	74.31	78.91	290.63	301.73	205.73	65.00	4.07
T_5	15.20	76.23	81.80	336.97	352.80	241.87	67.00	3.44
T_6	15.80	77.17	82.94	346.83	361.67	253.37	68.67	3.11
T_7	16.03	77.88	83.30	358.60	374.33	261.63	69.00	3.04
T_8	16.60	79.41	85.40	373.33	384.66	267.42	70.00	3.01
T_9	17.00	80.81	86.02	385.33	400.33	281.43	70.33	2.93
T_{10}	17.50	81.15	86.63	393.50	409.00	295.33	71.33	2.86
T_{11}	18.67	83.89	88.10	402.17	417.67	311.59	73.00	2.80
T_{12}	19.17	85.09	89.15	413.33	430.00	318.81	74.00	2.71
T_{13}	10.60	72.87	76.51	273.33	286.67	195.17	62.33	4.21
SEm±	0.56	1.24	1.41	15.54	15.51	13.15	2.71	0.82
CD (p=0.05)	1.33	2.97	3.39	37.26	37.19	31.53	6.50	NS

improves the metabolic activities of the plants (Chandra et al., 2015), thus could have aided in increased yield as higher quantum of carbohydrates and phyto-hormones are needed for fruit development and higher total yield (Sahu et al., 2015). Humic acid acts as chelating agent and improves chemical and microbial activity in the soil thus increases the availability of nutrients (Khattab et al., 2012) to the developing fruits. The improvement in fruit size, fruit and aril weight, fruit volume and juice content with the biostimulants application could also be attributed to sea weed extract, a component of biostimulant containing phyto-hormones and growth regulators like auxins, gibberellins, cytokinins (Panda et al., 2012), which individually or collectively might have improved the size and weight of fruits through cell division and enlargement (Taiz and Zeiger, 2006). Present findings are in conformity with those of Abubakar et al. (2013), who had recorded significantly higher yield, fruit size, weight and volume of pomegranate with the application of biostimulants. A significant increase in yield and fruit length as well as fruit weight of mango was reported with the application of sea weed extract by Mohamed and El-Sehrawy (2013) and with humic acid treatments by Ngullie et al. (2014).

Biostiumlants significantly improved bio-chemical fruit quality characteristics of pomegranate (Table 2). TSS, sugars and ascorbic acid contents increased but titratable acidity in the fruit juice decreased with the increasing doses of biostimulants. Highest TSS (15.27°B) as well as TSS: acid ratio (51.12) and lowest titratable acidity (0.30%) were recorded in fruits born on trees receiving T_{12} (Biostimulant B_1 @ 300 g plant^{-1} as two soil applications of 150 g each+two foliar sprays of Biostimulant B_2 @ 1.5 ml l^{-1}). This treatment was significantly superior to biostimulant B_1 @ 100 g and 200 g as single soil application (T_1 and T_2), biostimulant B_1 @ 100 g in two split doses (T_4) and control (T_{13}) with respect to TSS, whereas, with respect to titratable acidity it was statistically at par with all the treatments containing two split doses of biostimulant B_1+two foliar sprays of biostimulant B_2 (T_{10} and T_{11}). Treatment T_{12} was closely followed by T_{11} and was found significantly superior to all other treatments under study with respect to TSS: acid ratio. The maximum total sugars (13.55%), reducing sugars (10.40%) and ascorbic acid content (14.53 mg 100 g^{-1}) were recorded under treatment T_{12} (Biostimulant B_1 @ 300 g plant^{-1} as two soil applications of 150 g each+two foliar sprays of Biostimulant B_2 @ 1.5 ml l^{1}). This treatment was found significantly superior than those of treatments containing single dose of biostimulant B_1 (T_1 to T_3), two split doses of biostimulant B_1 @ 100 or 200 g (T_4 and T_5) and control (T_{13}). However, minimum TSS, TSS: acid ratio, total sugars, reducing sugars, ascorbic acid content and maximum titratable acidity were observed in fruits harvested from untreated (control) trees. The improvement in the bio-chemical fruit quality characteristics with the application of biostimulants may be ascribed to the presence of humic acid and seaweed extract in biostimulants. Humic

Table 2: Effect of biostimulants on fruit bio-chemical quality characteristics of pomegranate

Treatment	TSS (°B)	Titratable acidity (%)	TSS: acid ratio	Total sugars (%)	Reducing sugars (%)	Ascorbic acid (mg 100 g aril^{-1})
T_1	13.47	0.54	25.93	9.76	8.67	12.53
T_2	13.67	0.51	27.09	10.12	8.80	12.78
T_3	13.82	0.48	28.83	10.52	9.05	12.95
T_4	13.53	0.52	26.30	9.83	8.75	12.65
T_5	13.93	0.47	29.50	10.92	9.10	13.23
T_6	14.17	0.45	31.23	11.45	9.30	13.35
T_7	14.30	0.44	32.33	11.50	9.42	13.46
T_8	14.47	0.41	35.50	11.75	9.57	13.64
T_9	14.73	0.38	39.18	11.98	9.77	13.90
T_{10}	14.83	0.35	42.04	12.11	9.92	14.11
T_{11}	14.97	0.33	46.40	12.35	10.18	14.32
T_{12}	15.27	0.30	51.12	13.55	10.40	14.53
T_{13}	12.04	0.58	20.85	9.43	7.55	12.39
SEm±	0.65	0.03	2.31	0.91	0.53	0.49
CD (p=0.05)	1.55	0.07	5.54	2.18	1.28	1.18

acid is a constituent of organic matter in soils which has a relevant role in soil ecological functions and promotion of photosynthesis. Thus increase in TSS, sugars and ascorbic acid contents with the application of biostimulants might be attributed to increased photosynthetic efficiency and synthesis or accumulation of metabolites in the fruits. The results of present study are in line with the findings of Baksh et al. (2008), who had reported significant improvement in bio-chemical fruit quality characteristics (TSS, ascorbic acid and total sugars) of 'Sardar' guava with the application of biostimulants. Similar findings on improvement of bio-chemical fruit quality characteristics (TSS and sugars) have also been reported by Spinelli et al. (2009) in 'Fuji' apple, Ahmed et al. (2015) in mango and Rocha et al. (2016) in guava.

Chemical and biological properties (Table 3) were significantly influenced with different doses of biostimulants. Soil pH, electric conductivity and organic carbon did not show any consistent trend of increase or decrease with the increasing doses of biostimulants, however, available soil NPK contents and soil microbial count increased with the increasing doses of soil applied biostimulant B_1 but addition of foliar sprays of biostimulant B_2 failed to further increase the soil NPK contents and microbial count. The maximum soil pH (7.15) and organic carbon (0.96%) was recorded under treatment T_9 (Biostimulant B_1 @ 300 g as single soil application+one foliar spray biostimulant B_2 @ 1.5 ml l^{-1}) and this treatment was closely followed by treatment T_{12} (Biostimulant B_1 @ 300 g plant^{-1} as two soil applications of 150 g each+two foliar sprays of Biostimulant B_2 @ 1.5 ml l^{-1}). Electrical conductivity of soil was not influenced significantly by different doses of biostimulants. However, maximum available soil N (362.23 kg ha^{-1}), P (61.17 kg ha^{-1}) and K (310.12 kg ha^{-1}) were recorded with the treatment of biostimulant B_1 @ 300 g as single soil application+one foliar spray biostimulant B_2 @ 1.5 ml l^{-1} (T_9) which was closely followed by T_{12} (Biostimulant B_1 @ 300 g plant^{-1} as two soil applications of 150 g each+two foliar sprays of Biostimulant B_2 @ 1.5 ml l^{-1}). Both these treatments were found significantly superior to soil application of biostimulant B_1 @ 100 g in single and split doses (T_1 and T_4) and control (T_{13}). Minimum soil pH, organic carbon, available N, P and K were observed in untreated soil (control). The positive influence of biostimulants on soil chemical properties may be attributed to the constituents of biostimulant like sea weed extract and humic acid. Humic substances are most active component of soil and organic matter which is responsible for several complex chemical reactions in the soil (Trevisan et al., 2010), thus results in improving the availability and uptake of N, P and K contents. Humic substances are recognized as a key component of the soil fertility (Nardi et al., 2002) as it stimulates soil microbial activity, transform insoluble nutrients into soluble form and mobilizes them more effectively and thereby increase their availability to the plants. The results of present study are supported by the findings of Turan and Kose (2004) and Sangeetha et al. (2006) who reported an increase in nutrient uptake from soil with application of sea weed extract and

Table 3: Effect of biostimulants on chemical and biological properties of pomegranate orchard soil

Treatment	pH	Electrical conductivity (dSm^{-1})	Organic carbon (%)	Nitrogen ($kg\ ha^{-1}$)	Phosphorus ($kg\ ha^{-1}$)	Potassium ($kg\ ha^{-1}$)	Bacterial count ($\times 10^6$ cfu g^{-1})	Fungal count ($\times 10^4$ cfu g^{-1})	Actinomycetes count ($\times 10^4$cfu g^{-1})
T_1	6.93	0.197	0.88	329.72	42.78	276.61	35.33	6.00	11.67
T_2	6.96	0.195	0.89	342.52	48.26	283.93	36.67	6.67	12.67
T_3	6.98	0.197	0.91	353.24	53.23	292.94	37.33	7.33	13.67
T_4	6.94	0.194	0.88	337.37	44.19	279.60	36.00	6.33	12.00
T_5	6.99	0.198	0.92	351.19	50.42	289.56	38.67	7.00	12.67
T_6	7.03	0.200	0.93	357.01	56.13	303.30	40.33	8.00	14.33
T_7	6.95	0.203	0.90	339.23	44.80	291.51	35.67	6.67	12.33
T_8	7.05	0.204	0.94	357.46	56.57	303.65	42.00	8.33	14.67
T_9	7.15	0.205	0.96	362.23	61.17	310.12	44.67	10.33	16.33
T_{10}	6.96	0.205	0.90	345.35	47.66	295.73	37.67	6.67	13.00
T_{11}	7.08	0.207	0.94	357.83	53.20	296.60	39.33	7.67	15.33
T_{12}	7.11	0.212	0.95	360.27	58.84	306.25	42.67	8.67	15.67
T_{13}	6.90	0.190	0.85	317.92	36.06	266.70	30.67	4.67	10.33
SEm±	0.05	0.14	0.02	9.06	5.90	7.56	2.03	0.90	1.43
CD (p=0.05)	0.11	NS	0.06	21.71	14.14	18.11	4.87	2.15	3.42

Initial status of soil: pH-6.90, EC: 0.195 dSm^{-1}; Organic carbon: 0.82%; Available nitrogen: 314 kg ha^{-1}; Available phosphorus: 32.6 kg ha^{-1}; Available potassium: 258.40 kg ha^{-1}, Bacterial count: 29.33×10^6 cfu g^{-1}; Fungal count: 4.33×10^4 cfu g^{-1}; Actinomycetes count: 9.67×10^6 cfu g^{-1}

humic acid, respectively. A significant enhancement in uptake of macronutrients with the applications of humic acid was also reported by Jones et al. (2007) and Arjumend et al. (2015).

It is obvious from the data (Table 3) that biostimulants had a significant effect on microbial population of pomegranate orchard soil. Population of the bacteria, fungi and actinomycetes increased in the rhizosphere of pomegranate orchard soil with the increasing doses of biostimulants. Among the different treatments, maximum bacterial count (44.67×10^6 cfu g^{-1}), fungi population (10.33×10^4 cfu g^{-1}) and actinomycetes count (16.33×10^4 cfu g^{-1}) was recorded in the soil of tree basins received T_9 (biostimulant B_1 @ 300 g as single soil application+one foliar spray of biostimulant B_2 @ 1.5 ml l^{-1}) which was closely followed by T_{12} (Biostimulant B_1 @ 300 g $plant^{-1}$ as two soil applications of 150 g each+two foliar sprays of Biostimulant B_2 @ 1.5 ml l^{-1}). Both these treatments were found significantly superior to all the doses of biostimulant B_1 as single soil application (T_1 to T_3), soil application of biostimulant B_1 @ 100 g in two split doses (T_4), biostimulant B_1 @ 100 g in single soil application+one foliar spray of biostimulant B_2 @ 1.5 ml l^{-1} (T_7), soil application of biostimulant B_1 @ 100 g in two split doses+two foliar sprays of biostimulant B_2 @ 1.5 ml l^{-1} (T_{10}) and control (T_{13}) with respect to bacterial count, however, these were significantly super only to biostimulant B_1 @ 100 g as single soil application (T_1), soil application of biostimulant B_1 @ 100 g in two split doses (T_4), and control (T_{13}) with respect to soil fungi but significantly superior to biostimulant B_1 @ 100 g as single soil application (T_1), soil application of biostimulant B_1 @ 100 g in two split doses (T_4), biostimulant B_1 @ 100 g in single soil application+one foliar spray of biostimulant B_2 @ 1.5 ml l^{-1} (T_7) and control (T_{13}) with respect to actinomycetes population. The increase in soil microbial population with the use of biostimulants might be attributed to the seaweed extract and humic acid which improves soil aggregation, aeration, permeability, water holding capacity, hormonal activity, microbial growth, organic matter (Sarhan et al., 2011) and thereby the population of microbes in the soil through a congenial environment for their growth. The results of present study are in agreement with the findings of Sahain et al. (2007), who recorded an increase in the number of soil microflora (total fungi, bacteria and actinomycetes) with the biostimulant application to the apple orchard soil. Maji et al. (2016) also had reported a significant increase in bacterial and fungal diversity and density in soil supplemented with humic acid.

4. CONCLUSION

The treatment comprising granular biostimulant B_1 @ 300 g plant^{-1} as two soil applications of 150 g each+two foliar sprays of liquid biostimulant B_2 @ 1.5 ml l^{-1} was found to be the most effective combination of biostimulants for improving yield, fruit quality and bio-chemical soil properties of pomegranate.

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