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Effect of foliar spray of bio-inoculants and ammonium molybdate on growth and yield of broccoli (*Brassica oleracea* var. *italica*)

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Abstract
During the session 2021–2022, the experiment on "Effect of foliar spray of Bio-inoculants and Ammonium molybdate on growth and yield of Broccoli (*Brassica oleracea* var. *italica*)" was conducted at the Centre of Excellence on Protected Cultivation and Precision Farming (COEPCPF), while laboratory work was done at the Department of Agricultural Microbiology, IGKV, Raipur (Chhattisgarh), with the objective to see the effect of foliar spray of Bio-inoculants and Ammonium molybdate on Broccoli (TSX-2004). The experiment was laid out in Complete Randomized Design (CRD) with 7 treatments and 4 replications. Foliar spray of Bio-inoculants (*Azotobacter* + *Azospirillum*) at 30, 45 and 60 DAT and foliar application of Ammonium molybdate at three different concentrations (C) @ 0.10%, 0.20% and 0.40% solution was given after 06 days gap of spraying bio-inoculants. The experimental results revealed that significantly maximum plant height (28.5cm), number of leaves/ plant (21.8), total chlorophyll content (13.78 mg g^{-1} of leaves), dry shoot biomass (16.6 g/plant), dry root biomass (8.6 g/plant) were noted under T4 (IF + Foliar Application of Bio-inoculants and Ammonium molybdate 0.2%) at 60DAT, while minimum plant height (23.8 cm), number of leaves (16.0), total chlorophyll content (8.20 mg/g of leaves), dry shoot biomass (9.6 g/plant), dry root biomass (7.4 g/plant) were under T1 (IF). Treatments T4 (IF + Foliar Application of Bio-inoculants and Ammonium molybdate 0.2%), and T3 (IF + Foliar Application of Bio-inoculants and Ammonium molybdate 0.1%) were recorded significantly highest yield attributes *viz*. fruit diameter and fruit weight per plant (359.0 g and 349.8 g) which proved the best treatment combination in comparison to other treatments.

Keywords: Broccoli, bio-inoculants, micronutrients, growth and yield

Introduction
Broccoli is an Italian word that comes from the Latin word *branchium*, which means "arm" or "branch" (Ujjwal *et al*., 2020) [18]. Broccoli (*Brassica Oleracea* L. var. *italica*) is a Cruciferae vegetable. The broccoli plant morphologically resembles cauliflower and is known in the local language as 'Hari Gobi,' which means green cauliflower. Broccoli has 14 times the amount of beta-carotene, a vitamin A precursor, as regular cabbage (Sharma, 2000) [11]. It is an important vegetable because it contains vitamins, antioxidants, glucosinolates, and other anti-cancer chemicals (Parente *et al.*, 2013) [8]. 100 g of broccoli include 7 g of carbohydrate, 0.4 g of fat, 33 mg of sodium, 316 mg of potassium, 2.6 g of dietary fibre, 1.7 g of sugar, and 2.8 g of protein. Vitamin A (12 IU), Vitamin C (148 mg), Calcium, Iron, Vitamin B6, Magnesium, and Phosphorus are all included (Annon., 2015) [1]. Although broccoli is an under-utilized food crop in India, it is confined to a small area, particularly near major towns. Broccoli is commercially grown in Himachal Pradesh, Uttar Pradesh, Jammu & Kashmir, the Nilgiri Hills, and India's northern plains (Singh *et al*., 2021) [16].

Azotobactor is a bio-inoculant that works in a non-symbiotic relationship with non-leguminous crops to fix nitrogen in the environment. *Azotobacter* inoculation saves roughly 20 to 40 kg nitrogen per hectare. The beneficial effects of *A. chroococcum* are attributed to production of plant growth hormones, improved nutrient uptake and antagonistic effect on plant pathogens (Parmar and Dadarwal, 1997) [9].

In plants, molybdenum is known to act as a catalyst for enzyme activity. Plants can get it as the $HMoO_4^-$ ion. Apart from the primary nutrients, molybdenum (Mo) is important micronutrients for optimal growth and development in Cole crops, (Shelp and Liu, 1992) [13].This deficiency causes many anatomical, physiological and biological disorders (Shelp and Liu, 1992) [13].

Foliar application may thus be a promising strategy for increasing broccoli growth and yield. Molybdenum (Mo) helps in N2 fixation because it is a part of metaprotein nitrogenase (Gupta and Vyas, 1994) [2]. Molybdenum is also an essential micronutrient in symbiotic nitrogen fixation. Hence the study is being proposed to see the effect of foliar spray of bio-fertilizer and micronutrient on performance of Broccoli.

Materials and Methods

The investigation entitled "Effect of foliar spray of Bio-inoculants and Ammonium molybdate on growth and yield of Broccoli (*Brassica oleracea* var. *italica*)" was conducted during 2021-2022 under poly house condition at the Centre of Excellence on Protected Cultivation and Precision Farming (COEPCPF) and laboratory work was carried out in the Department of Agricultural Microbiology, College of Agriculture, Raipur, Chhattisgarh to assess the effect of Bio-inoculants and Ammonium molybdate on growth and yield of Broccoli (TSX-2004). The location is characterized by sub-humid climate with average rainfall of 1200-1400mm concentrated during rainy season (June –September) which is situated in plains of Chhattisgarh at latitude 21°16'N and longitude 81°36'E with an altitude of 289.60 m above mean sea level (MSL). The experiment was laid out in Complete Randomized Design (CRD) with 7 treatments and 4 replications. Treatments consisted of T1 (IF), T2 (IF + Foliar spray of Bio-inoculants), T3 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.1%), T4 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.2%), T5 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.4%), T6 (IF + Foliar spray of Ammonium molybdate 0.2%) and T7 (IF + Foliar spray of Ammonium molybdate 0.4%). Foliar spray of Bio-inoculants (*Azotobacter* + *Azospirillum*) at 30, 45 and 60 DAT and foliar application of Ammonium molybdate at three different concentrations (C) @ 0.10%, 0.20% and 0.40% solution was given after 06 days gap of spraying bio-inoculants. Spraying was done with the help of hand sprayer. Observations on plant height (cm), Number of leaves/plant, total chlorophyll content, Curd diameter (cm), Weight per fruit (g) and dry biomass of shoot and root (g) were recorded by randomly selecting four plants from each plot.

1. Plant Height (cm)

From each treatment, four plants were randomly chosen and permanently labeled. Each tagged plant's height was measured on a metre scale from the base to the tip of the main shoot at 15, 30, 45 and 60days of transplanting with the average height of four plants recorded as mean plant height in centimetre.

2. Number of leaves per plant

Except for leaves adhering to the heads, all fully grown leaves were counted. At 15, 30, 45 and 60 days of transplanting, the number of leaves per plant of randomly selected plants from each treatment and replication were counted, and the mean number of leaves per plant was calculated.

3. Total Chlorophyll Content in leaves at curd initiation stage

The chlorophyll content of leaves at curd initiation stage was determined by Acetone Method at 50 days after transplanting. 0.2g of fresh plant leaf material solubulising with 25 ml of 80% acetone and the optical density of the extract is measured at 663 and 645 nm wavelengths using spectrophotometer because at these wavelengths, maximum absorption of chlorophyll "a" and "b" takes place, respectively.

The total chlorophyll was calculated using the formula.

Mg total chlorophyll/g tissue = $20.2(A_{645}) + 8.02(A_{663}) \times V1000/W$

Where

A= absorbance at specific wavelengths

V= final volume of chlorophyll extract

W= fresh weigh of tissue extracted

4. Curd diameter (cm)

The curd diameter was calculated as the average of the 4 random marketable curds measured by the help of a thread and scale at the widest regions of the curd.

5. Weight per fruit (g)

At harvest, all fruits from each treatment and replication were weighed. Its weight was recorded, and the average value was calculated.

6. Dry biomass of shoot and root (g/plant)

The plant parts like shoot and roots were collected from the growing test plants while sampling at harvest. These samples were then oven dried at 65°C till it reached at steady weight for 3-5 days. The final dry weight of the shoot and roots were observed and recorded according to their corresponding treatments.

Statistical analysis

The data on growth and yield were subjected to the Panse and Shukhatme method of analysis of variance (ANOVA), where the F tests was significant for comparison of the treatment means, CD values were worked out at 5% probability level.

Result and Discussion

Plant Height (cm)

The treatment T4 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.2%) recorded significantly maximum (28.5cm) height of plant on 60DAT, which was at par with T3. Whereas significantly minimum height of plant (23.8 cm) was recorded in the treatment T1 (IF). Similar results were obtained by Singh *et al.*, (2018) [15].

Number of leaves per plant

The maximum number of leaves per plant was observed under the treatment of T4 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.2%) at curd formation stage (21.8), which was significantly different from T3 (19.8) followed by T2 (18.3) and T6 (17.8). The minimum number of leaves per plant was seen at T1 (IF), (16.0) at 60 DAT. The result corroborated with the findings of Pankaj *et al.*, (2018) [10] and Kumar *et al.*, (2011) [11] on cauliflower (cv. pusa snowball k-1).

Total Chlorophyll Content in leaves at curd initiation stage

The IF + Bio-inoculants + Ammonium molybdate significantly affected the total chlorophyll content. The maximum total chlorophyll content (13.78 mg/g) was recorded under T4 (IF + Foliar spray of Bio inoculants and

Ammonium molybdate 0.2%) while minimum (8.20mg/g) chlorophyll content was recorded under T1 (IF). Similar results were obtained by Thapa *et al*., (2016) [17].

Curd diameter (cm)

The treatment T4 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.2%) recorded maximum curd diameter (22.9 cm), however treatment T1 (IF) recorded minimum curd diameter (18.8 cm). Similar result obtained by Pushpanjali *et al*., (2018).

Weight per fruit (g)

The treatment T4 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.2%) recorded the maximum fruit yield (359.0 g) which was at par with T3 (349.8 g) but significantly differ from T2 (343.0 g) and other treatments. The significantly minimum fruit yield/plant was recorded in treatment T1 (IF) (268.0 g). Similar result obtained by Mohamed and Abdelnaser (2011) [6] on cauliflower, Khare and Singh (2008) [3] on cabbage cv. Golden acre and Sharma *et al.*, (2002) [12] on cabbage.

Dry biomass of shoot and root (g/plant)

The maximum shoot dry weight was recorded in T4 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.2%) (16.6 g/plant) followed by T3 and T2 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.1% and IF + Bio-inoculants) (14.5 g/plant and 13.6 g/plant). Whereas maximum root dry weight was recorded in T4 (IF + Foliar spray of Bio-inoculants and Ammonium molybdate 0.2%), (8.6 g/plant) followed by T3 (8.2 g/plant) and T2 (8.1 g/plant). Biomass accumulation in broccoli plant at harvest was found significantly varied among treatments, being highest with application of bio-inoculants and spray of Molybdenum at moderate dose @ 0.2%. Similar result obtained by Manivannan and Singh (2004) [5] and Singh *et al.*, (2017) [14].

Table 1: Effect of Bio-inoculants and Ammonium molybdate on plant height and number of leaves of broccoli at different growth stages

Treatments	Treatment details	Plant height (in cm)				No of leaves per plant			
		15 DAT	30 DAT	45 DAT	60 DAT	15 DAT	30 DAT	45 DAT	60 DAT
T1	IF	7.6	12.2	17.8	23.8	5.0	7.3	11.0	16.0
T2	IF + Foliar Application of Bio-inoculants	8.7	13.5	19.2	24.4	5.5	7.8	12.3	18.3
T3	IF +Foliar Application of Bio-inoculants and Ammonium molybdate 0.1%	9.4	13.8	22.8	26.6	5.3	8.5	13.3	19.8
T4	IF +Foliar Application of Bio-inoculants and Ammonium molybdate 0.2%	9.7	15.3	23.6	28.5	5.3	9.0	13.3	21.8
T5	IF +Foliar Application of Bio-inoculants and Ammonium molybdate 0.4%	8.6	14.2	20.0	24.0	5.0	7.0	12.5	17.3
T6	IF +Foliar Application of Ammonium molybdate 0.2%	9.0	14.5	20.5	24.5	5.3	7.8	13.5	17.8
T7	IF +Foliar Application of Ammonium molybdate 0.4%	8.9	13.5	20.0	23.8	5.3	8.3	11.5	16.8
SEm±		NS	NS	0.361	0.683	NS	NS	0.332	0.469
CD at 0.05%		NS	NS	1.069	2.022	NS	NS	0.983	1.390

Table 2: Effect of Bio-inoculants and Ammonium molybdate on Chlorophyll content, yield attributes and biomass accumulation in Broccoli crop at harvest

Treatments	Treatment details	Chlorophyll content (mg/gLFW)	Curd diameter (in cm)/one curd	Weight per fruit (in g)	Dry biomass (in g/plant)	
					Shoot	Root
T1	IF	8.20	18.8	268.0	9.6	7.4
T2	IF + Foliar Application of Bio-inoculants	12.45	19.8	343.0	13.6	8.1
T3	IF +Foliar Application of Bio-inoculants and Ammonium molybdate 0.1%	13.24	21.4	349.8	14.5	8.2
T4	IF +Foliar Application of Bio-inoculants and Ammonium molybdate 0.2%	13.78	22.9	359.0	16.6	8.6
T5	IF +Foliar Application of Bio-inoculants and Ammonium molybdate 0.4%	13.23	19.5	298.3	12.9	7.8
T6	IF +Foliar Application of Ammonium molybdate 0.2%	11.23	19.5	318.0	13.2	7.7
T7	IF +Foliar Application of Ammonium molybdate 0.4%	10.54	19.2	282.8	12.5	7.3
SEm±		0.211	-	5.603	0.284	0.146
CD at 0.05%		0.625	-	16.591	0.839	0.434

Note

- IF –Inorganic fertilization (50% RDF) through Fertigation
- Bio-inoculants: Foliar spray (*Azotobacter +Azospirillum)* at 30, 45 and 60 DAT.
- Foliar Application of Ammonium molybdate was given after 06 days gap of spraying bio-inoculants.

Conclusion

Broccoli plant cv. (TSX-2004) responded well as per growth and yield considered to treatment T4 and T3 consisting of foliar application of bio-inoculants and lower doses of micronutrient (0.1% and 0.2%), i.e., Ammonium molybdate. Under treatment, T4, we recorded maximum plant height (28.5cm), no. of leaves (21.8), total chlorophyll content (13.78mg/g), dry biomass of shoot (16.6g) and root (8.6g), curd diameter (22.9cm) and curd weight (359 g), as compared to other treatments. It can be recommended for commercial production of broccoli to enhance the yield and quality of fruit.

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