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Effect of microbial inoculants on growth and yield of chilli (*Capsicum annum* L.) under moisture deficit conditions

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Abstract

A pot experiment entitled "Effect of microbial inoculants on growth and yield of chilli (*Capsicum annum* L.) under moisture deficit conditions." was carried out in the Division of Basic Sciences & Humanities Faculty of Horticulture SKUAST-K to evaluate the role of individual and combined applications of different microbial strains like AM fungus, PSB, KSB and ZnSB in chilli under moisture deficit conditions (MDC) (60% FC). The results of the study revealed that growth and yield parameters of chilli showed a significant reduction under MDC (T1) as compared to control (T0) (100% FC). Inoculation with different microbial strains significantly improved all the examined parameters in chilli as compared to control as well as MDC. Among the different microbial strains used in the experiment, the combined application of AM, PSB, KSB and ZnSB (T6) proved to be the best in terms of significantly enhancing most of the studied parameters of chilli as compared to control or MDC. Among the parameters that enhanced due to combined inoculation of microbial strains plant height, root biomass, total plant dry weight, total leaf area, number of fruits, fruit weight and yield showed a respective increase of 18.8, 69.6, 77.3, 34.0, 67.0, 48.2, 82.91, as compared to T1.

Keywords: Chilli, growth, microbial inoculants, moisture deficit

Introduction

Chilli (*Capsicum annum* L.) is one of the most important vegetable crops grown for its fruit. It belongs to the family Solanaceae and genus Capsicum. Chilli is native to Mexico, Southern Peru and Bolivia (Villalon, 1981)^[20] where it is used as one of the main spice ingredients. In India it was introduced by Portuguese towards the later part of 15^{th} century. There are mainly five cultivated Capsicum species which are *C. Annuum*, *C. baccatum*, *C. chinense*, *C. frutescens and C. pubescens*. Among these *C. Annuum* L. is the most widely cultivated species grown for its pungent and non pungent fruits which are respectively called as hot pepper and sweet pepper (Bosland and Votava, 2000)^[6]. Chilli is a rich source of vitamin 'C', vitamin A, minerals, antioxidants, carotene and zeaxanthin that are known to have disease preventing and health promoting properties. Some varieties of chilli are famous for red colour because of the pigment capsanthin where as others are known for biting pungency attributed to capsaicin (Macrae *et al.*, 1993)^[13]. Capsaicin has anti-cancerous properties and it is also used for preparing drugs for heart disease and cosmetics.

Chilli is widely cultivated throughout the warm, temperate, tropical and subtropical countries because it requires long season and warm climate for its best growth and development. Chilli crop is raised over an area of 1832 thousand hectares globally with an annual production of 2959 thousand tonnes (Anonymous, 2009)^[1]. India contributes one fourth of world production of chilli with an average annual production of 18.72 lakh tonnes from an area of 8.30 lakh hectares (Anonymous, 2017)^[3].

The term bio-fertilizer is generally defined as a preparation containing live or latent cells of efficient strains of N-fixing, P solubilizing or cellulolytic microorganisms used for seed or soil application. Mixed inoculants interact additionally to perform better and show quick results. Many researchers have evidenced that development of plant growth promoting consortium, could be a feasible technology for increasing the effect of microbial inoculants on chilli (Anonymous, 2011)^[2].

Since a lot of work has been carried out on the application of bio fertilizers but their combined role in relation to the water scarcity needs much attention especially in rainfed areas of India and J&K. Hence, an attempt has been made in this research to assess the role of individual and combined application of bio fertilizers in chilli under moisture deficit conditions on its growth and yield.

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Materials and Methods Pot filling

The pots of uniform size (27×24.5cm) and weight (0.370 kg) were filled with 8.0 kg of soil collected from experimental field of Division of vegetable Science, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shalimar, Srinagar. Soil was mixed with farm yard manure in a ratio of 2:1. Little amount of sand was mixed thoroughly in each pot to improve aeration.

Crop variety

The variety used in the experiment was Kashmir long-1.

Seedling source

The seedlings of chilli Kashmir long-1 were obtained from the Division of Vegetable Science, Sher-e-Kashmir University of Agricultural Sciences and Technology of Kashmir, Shalimar, Srinagar.

Treatment details

The treatments used for the current study were T0 Control (100% field capacity), T1 MDC (Moisture deficit condition) [60% field capacity], T2 MDC + AM (Arbascular mycorrhiza), T3 MDC + AM+PSB (Phosphate solubilising bacteria), T4 MDC + AM+KSB (Potassium solubilising bacteria), T5 MDC + AM + ZnSB (Zinc solubilising bacteria) and T6 MDC + AM + PSB + KSB + ZnSB.

Creation of moisture deficit conditions

After filling the pots with 8.0 Kg of soil, pots selected as T0 (100% FC) were flood irrigated followed by weighing when the movement of gravitational water stopped. The increase in weight of the pots was representative of the water held by the soil. In our case the water absorbed by 8.0 kg of dried soil was 2.30 kg (2.30 lts) which means that 2.30 kg of water brought the pots to 100% field capacity. In all other pots where FC was to be maintained at 60%, 1.38 kg (1.38 lts) (60% of 2.30 Kg) of water was maintained by irrigation. The field capacity was maintained at 100% for T0 and 60% for all other treatments by measuring the weight of pots daily and subsequently adding the water equal to the loss in the pot weight. The pots were shifted to glass house during night to avoid the rains.

Application of AM

AM @20 g per pot was applied near the root zone of the plants before transplanting of seedlings.

Application of PSB, KSB and ZnSB

The roots of the seedlings were dipped for 20 minutes respectively in the solution of PSB, KSB and ZnSB @ 20 ml after which seedlings were dried in shade for 30 minutes till the white film was visible over the root surface of seedlings. However, combined inoculation (T6) was prepared by mixing 7 ml of PSB, KSB and ZnSB to form a consortium (approx. 20 ml) and seedlings were dipped in this consortium for 20 minutes followed by drying in shade for 30 minutes before transplanting.

Transplanting of seedlings

After applying the treatments, seedlings were transplanted into the pots. Initially three seedlings were transplanted in each pot till their establishment and thereafter only one seedling was left in the pot and rest of the two were thinned out. Parameters recorded were Plant height (cm), root biomass (g/ plant), plant dry weight (g/ plant), total leaf area (cm2/ plant), number of fruits (per plant), fruit weight/ plant (g) and yield per plant (g).

Design of the experiment

The design adopted for the experiment was Completely Randomized Design (CRD) as described by Gomez and Gomez (1984)^[23] and critical difference (CD) values were calculated at 0.05% level of significance.

Results and discussion Results

In this experiment it was observed that at T1 the plant height of chilli was significantly reduced by 12.5% as compared to T0 (table-1). With the incorporation of AM fungus in the growing medium (T2), the plant height of chilli was significantly increased as compared to T0 as well as T1. Plant height at T2 showed a respective increase of 4.7% and 9.8% as compared to T0 and T1. Inoculation of chilli seedlings with AM and PSB (T3) enhanced the plant height by 10.5% and 13.5% as compared to T0 and T1. Similarly in case of seedlings, inoculated with AM and KSB (T4), there was an increase of 9.1% and 14.1% as compared to T0 and T1 in plant height which was statistically at par with T3. Inoculation of chilli seedlings with AM and ZnSB (T5) also showed the similar type of results where the plant height was increased by 6.4% and 11.4% as compared to T0 and T1. However, the combined inoculation with all the microbes under MDC proved to be best in terms of increasing the plant height. It can clearly be seen from that plant height at T6 increased by 14.3% as compared to T0, where as it was increased by 18.8% as compared to T1.

From table-1 it is evident that at T1 the root biomass of chilli was decreased in comparison to T0 by 23.6%. Inoculation with AM to chilli seedlings growing under MDC (T2) showed a positive effect where the root biomass was increased by 50.6% and 62.3% as compared to T0 and T1. The dual inoculation of moisture stressed seedlings with AM in addition to PSB (T3), KSB (T4) and ZnSB (T5) also showed an increasing effect on root biomass accumulation. Root biomass at T3 was increased by 58.6% and 68.3% as compared to T0 and T1. Similarly in case of T4 root biomass was enhanced by 57.3% and 67.4% as compared to T0 and T1.At T5 the increment in root biomass was estimated 54.1% and 64.9% over T0 and T1. However, the inoculation with all the soil microbes used in this study showed the best result in enhancing the root biomass of chilli. It is clear from that root biomass at T6 increased by 60.3% and 69.6% as compared to T0 and T1 respectively.

The data pertaining to the total plant dry weight as shown in table-2 indicates that at T1 the total plant dry weight of chilli was significantly reduced by 44.9% as compared to T0. However, when the seedlings subjected to MDC were inoculated with AM fungus (T2), it was found that total plant dry weight of chilli was increased by 6.4% and 61.1% as compared to T0 and T1. Similarly, inoculation of seedlings with AM and PSB (T3), AM and KSB (T4) and AM and ZnSB (T5) under MDC had a positive effect on the plant dry weight as compared to un-inoculated chilli seedlings subjected to MDC. Total plant dry weight at T3 was increased by 29.3% and 61.1% as compared to T0 and T1. Similarly at T4 an increase of 24.3% and 58.3% and at T5 an increase of 58.7% and 77.2% was observed over T0 and T1 respectively. Combined inoculation of seedlings with all the microbial

strains under MDC (T6) proved best in terms of increasing the plant dry weight as compared to T0 and T1 respectively by 58.7% and 77.3% as indicated in table-2.

It is obvious from table-2 that total leaf area of chilli was reduced by subjecting the un-inoculated chilli seedlings to MDC (T1) by 15.4% as compared to T0. Inoculation of moisture stressed seedlings with AM fungus (T2) recorded an increase in total leaf area as compared to T0 and T1by 0.21% and 15.5%. Similarly, as compared toT0 and T1 an increment of 13.0% and 26.3% was observed in total leaf area at T3. At T4 and T5 total leaf area of chilli was increased by 9.2% and 0.52% as compared to C0 r11 respectively. However, in case of T6 where the moisture stressed seedlings have been inoculated with all the microbial strains, total leaf area of chilli plants was increased to maximum. It can clearly be seen from table-2 that total leaf area of chilli at T6 was increased by 22.1% and 34.0% as compared to T0 and T1 respectively.

Number of fruits per plant recorded a significant reduction as the plants were subjected to MDC (T1). It can clearly be seen from table-3 that number of fruits/plant at T1 was reduced by 36.9% as compared to T0. Number of fruits/plant recorded an increase by inoculating the moisture stressed seedlings with AM fungus (T2). At T2 the number of fruits/plant was increased by 37.0% and 60.3% as compared to T0 and T1.Similarly, as compared to T0 and T1 an increase of 45.9% and 65.9% was observed in number of fruits/plant at T3. At T4 the percent increase in number of fruits/plant was recorded to be 42.2% and 63.6% over T0 and T1 followed by an increase of 38.3% and 61.1% at T5. However, among all the treatments number of fruits/plant was maximum when the seedlings growing under MDC were inoculated with all the microbial strains (T6). It is clear from table-3 that number of fruits/plant was increased by 47.6% and 67.0 as compared to T0 and T1 respectively.

Fruit weight in seedlings subjected to MDC (T1) observed a decrease as compared to T0. As, can be seen from table-3 that fruit weight in chilli at T1 was significantly reduced by 26.3% as compared to T0. Inoculating the moisture stressed seedlings with AM (T2) enhanced the fruit weight by 7.4% and 31.7% as compared to T0 and T1 respectively. In combination with PSB, AM (T3) fungus proved better in increasing the fruit weight which increased by 23.6% and 43.6% as compared to T0 and T1. Similarly, AM with KSB (T4) and AM with ZnSB (T5) had a positive effect in increasing the fruit weight of chilli subjected to MDC. Fruit weight at T4 was increased by 22.1% and 42.5% followed by an increase of 11.2% and 34.5% at T5 respectively as compared to control T0 and T1. Maximum fruit weight was obtained when a combined inoculation of moisture stressed seedlings was made. It can be seen from table-3 that at T6 fruit weight was increased by 48.2% and 29.7% as compared to T1 and T0 which was more as compared to rest of the treatments used in the study.

The yield per plant under MDC (T1) reduced as compared to T0 by 53.5% as depicted in table-3. The inoculation of chilli seedlings with AM increased the yield/ plant by 41.71% and 72.91% as compared to T0 and T1 respectively. The inoculation of chilli seedlings with AM+PSB (T3), AM+KSB (T4), AM+ZnSB (T5) and AM+PSB+KSB+ZnSB (T6) further showed an increment in yield/ plant by 58.69%, 55.04%, 45.35% and 63.23% as compared to T0 and by 80.80%, 79.10%, 74.60% and 82.90% as compared to T1. Thus it can be concluded that at T6 the yield/ plant was found highest as compared to rest of the treatments (table-3).

Table 1: Effect of Microbial inoculants on plant height and root

 biomass of chilli (Kashmir long-1) under moisture deficit conditions

Treatments	Plant height	Root
	(cm)	Biomass (g)
T0 : Control	49.07	0.72
T1: MDC	46.43	0.55
T2: MDC+ VAM	51.53	1.57
T3 : MDC+ VAM + PSB	54.83	1.74
T4 : MDC+ VAM + KSB	54.00	1.69
T5 : MDC+ VAM + ZNSB	52.43	1.46
T6 : MDC+ VAM +PSB +KSB+ ZNSB	57.23	1.81
C.D (p≤0.05)	1.32	0.087

 Table 2: Effect of microbial inoculants on total plant dry weight and total leaf area of chilli (Kashmir long- I) under moisture deficit conditions

Treatments	Total plant dry weight (g)	Total Leaf area/plant (cm ²)
T0 : Control	4.36	1,696.47
T1: MDC	2.40	1,436.30
T2: MDC+ VAM	4.66	1,700.17
T3 : MDC+ VAM + PSB	6.17	1,950.40
T4 : MDC+ VAM + KSB	5.76	1,869.80
T5 : MDC+ VAM + ZNSB	5.29	1,705.00
T6 : MDC+ VAM +PSB +KSB+ ZNSB	10.56	2,177.80
C.D (p≤0.05)	0.28	69.81

 Table 3: Effect of microbial inoculants on number of fruits/plant

 fruit weight and yield/plant of chilli (Kashmir long-1) under

 moisture deficit conditions

Treatments	Number of fruits per plant	Fruit Weight	Yield/ plant (g)
T0 : Control	20.47	4.72	96.61
T1: MDC	12.90	3.48	44.89
T2: MDC+ VAM	32.50	5.10	165.75
T3 : MDC+ VAM + PSB	37.85	6.18	233.91
T4 : MDC+ VAM + KSB	35.46	6.06	214.88
T5 : MDC+ VAM + ZNSB	33.23	5.32	176.78
T6 : MDC+ VAM +PSB +KSB+ ZNSB	39.10	6.72	262.75
C.D (p≤0.05)	1.76	0.47	0.065



Fig 1: Transplanting of chilli seedlings



Fig 2: Estimation of total leaf area in chilli

Discussion

The experimental findings reveal that all the growth parameters like plant height, root biomass, plant dry weight and number of leaves in chilli significantly reduced by subjecting the seedlings to MDC which respectively recorded a reduction of 12.5%, 23.6%, 44.9% and 15.5% as compared to control. These results are in agreement with that of Shintu and Jayaram (2015)^[19] who reported that the examined growth parameters of tomato significantly reduced as the seedlings were subjected to MDC as compared to control.

Reduction in plant height of chilli growing under MDC can be attributed to limited water availability which might have reduced the cell turgor pressure responsible for cell elongation as evident from our results on cell relative water content (table-1). Plant height of chilli seedlings was significantly increased by inoculating the seedlings with different microbial inoculants as compared to height of plants subjected to MDC. Ruiz-Lozano et al. (1995) [17] have also reported that inoculation with AM to lettuce has consistent effects on plant growth parameters under MDC. The positive effect of AM on plant growth can be attributed to increase in root surface area and its ability to extract the moisture from deeper layers of soil hence, increasing the capacity to absorb the water under MDC as evident from the increased leaf relative water content of chilli in our study. The increase in plant height at T3, T4, T5 and T6 can be contributed to solubilisation of phosphorus, potassium and zinc by inoculation with PSB, KSB and ZnSB followed by their consequent uptake by AM which otherwise, become insoluble and unavailable to plants under MDC. Same results have also been found by Bashir et al. (2017)^[5] and Shafi et al. (2019) ^[19] who reported an increase in plant height of chilli by inoculation of seedlings with PSB and KSB through rapid solubilisation of essential mineral nutrients through organic acid production.

It was found that root biomass decreased by 23.6% as compared to control by subjecting the seedlings to MDC. Similar results have also been found by Farooq *et al.* (2009)^[9] and Budayati *et al.* (2017)^[7] who reported that scarcity of water is a severe environmental constraint to plant growth and productivity. Drought stress reduces leaf size, stem extension and root proliferation, which disturbs plant water relations

and reduces water-use efficiency. The lesser root biomass in chilli under MDC can be attributed to reduced root length due to water deficiency and lesser availability of nutrients particularly the P which has a major role in root growth. It has also been reported that root meristimatic activity is highly susceptible to attack of free radicals which are overproduced in drought sensitive species because of reduction in the total antioxidant activity hence inhibiting the root growth (Gill and Tuteja, 2010)^[10].

The root biomass was enhanced when chilli seedlings were inoculated with AM fungus. Similar findings were revealed by Huixing (2005)^[11] who reported that AM improves the properties of soil in rhizosphere and enlarges root area of host plants. Besides AM fungus maintains the cell turgor pressure even under drought conditions, hence promoting the root growth by minimizing its effect on the rate of cell expansion. The application of AM+PSB, AM+KSB and AM+ZnSB under MDC further increased the root biomass of chilli as compared to AM alone. Jalal *et al.* (2014)^[13] also reported that Pseudomonas increased root length, root and shoot dry weight significantly.

The total plant dry weight decreased under MDC in comparison to control by 44.9%. The decrease in total plant dry weight under MDC was also found by Ibrahim *et al.* (2014)^[12] who reported that water stress significantly affected total dry weight accumulation in apple.

The inoculation of chilli seedlings with AM fungus under MDC led to a gain in total plant dry weight of chilli. The higher dry weight in plants inoculated with AM could be ascribed to its ability to increase the leaf relative water content and leaf water potential. The increase in plant dry weight through dual or combined inoculation of plants with AM and other soil microbes can be ascribed to the ability of these soil microbes to solubilise the fixed phosphorus, potassium and zinc by secreting the organic acids followed by their uptake and taking part in the different plant physiological processes and hence contributing to gain in total plant dry weight under MDC.

Total leaf area per plant was reduced under MDC by 15.4% as compared to control. The reduction in total leaf area under MDC was also studied by Widuri et al. (2017) [22] who reported that leaf is a sensitive plant organ in responding to abiotic stresses, especially drought stress which inhibits the leaf expansion in chili pepper hence affecting its total leaf area. The inoculation with AM fungus enhanced the total leaf area in chilli seedlings as compared to control. Similar findings were reported by Sandhiya et al. (2013) [18] who studied the positive effects of AM on various growth parameters including the total leaf area. The inoculation of chilli seedlings with AM+PSB, AM+KSB and AM+ZnSB under MDC further enhanced the total leaf area. The combined inoculation with all the microbial strains under MDC yielded maximum total leaf area as compared to rest of the treatments. These findings are in close conformity with those of Sandhiya et al. (2013) ^[18] who reported that total leaf area/plant is maximum with dual inoculation of soil microbes than their individual inoculation.

The number of fruits per plant reduced significantly under MDC as compared to control by 36.9%. The reduction in number of fruits per plant under deficit irrigation was also reported by Dorji *et al.* (2005) ^[8] who reported that fruit number per plant was reduced by more than 20% in deficit irrigation compared to normal irrigation. The decrease in number of fruits per plant under MDC can be attributed to more sensitivity of crops towards drought at flowering stage

which lead to high floral abortion ultimately affecting fruit number (Katerji et al., 1992)^[14]. The addition of AM fungus significantly increased the fruit number in chilli as compared to MDC (T1). Similar results were also found for rest of the treatments where the plants subjected to MDC where inoculated with AM and other biofertilizers like PSB, KSB and ZnSB in duplicate or in combination. Same results were found by Shintu and Jayaram (2015) [19] and Pandiyarajan (1995)^[16] who respectively reported increase number of fruits in tomato and onion when primed with different biofertilizers. The increased fruit number under such conditions may be due to maintenance of plant water relations hence, avoiding the damage caused because of low water availability at flowering stage. Besides, availability of essential elements like P, K and Zn is highly enhanced because of PSB, KSB and ZnSB which otherwise become limiting for the proper fruit growth and development. The fruit weight was negatively influenced under MDC by 26.3% as compared to control. The reduction in fruit weight under deficit irrigation was also studied by Dorji et al. (2005)^[8] who reported that total fresh mass of fruit was reduced by 34.7% in deficit irrigation compared to normal irrigation. The reduction in fruit weight per plant under MDC has been considered to be because of reduced photosynthetic rate. As a consequence water stress decreases the amount of phtosynthates exported from the leaves (Dorji et al., 2005) [8]. The fruit weight under MDC was increased when seedlings were inoculated with AM fungus as compared to control. Same results were achieved in drought stressed seedlings when they were inoculated with AM, PSB, KSB and ZnSB in duo or a combination of all the soil microbes. These results are in conformity with Ayub et al. (2000)^[4] who studied the effect of AM on drought tolerance and growth of plants and reported that inoculation with AM increased the fruit weight per plant. As has been reported by several workers that AM enhances the water uptake even under drought conditions hence plants experience a negligible effect on the photosynthetic rate as has been proved in our study also. Besides the nutrient solubilizers applied release the insoluble P, K and Zn and their uptake is enhanced by AM fungi which in turn promote the plant and fruit growth.

Conclusion

In the light of the experimental findings, it is concluded that moisture deficit adversely affected all the selected growth and vield parameters. Inoculation of moisture stressed chilli seedlings (var. Kashmir long-1) with different microbial strains viz; AM, PSB, KSB and ZnSB either alone or in combination improved growth and yield characteristics in chilli as compared to control and un-inoculated seedlings growing under moisture deficit conditions. Among the microbial inoculants, the combined application of chilli with VAM + PSB + KSB + ZnSB under moisture deficit conditions proved to be the best in terms of enhancing growth and yield attributes of chilli as compared to individual or dual application of inoculants. Therefore from our findings it can be concluded that the combined application of different strains of microbial inoculants can alleviate adverse effects of moisture deficit in chilli as compared to their individual application.

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