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Growth Response of Capsicum (*Capsicum annuum* **L.) to Varying Levels of Phosphorus, Zinc and Arbuscular Mycorrhizae**

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Authors' contributions

This work was carried out in collaboration among all authors. Author GB designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Authors US and PSB managed the analyses of the study. Author RK managed the literature searches. All authors read and approved the final manuscript.

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Original Research Article

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ABSTRACT

The present study was conducted to evaluate the growth response of Capsicum (*Capsicum annuum* L.) to varying levels of Phosphorus (P), Zinc (Zn) and Arbuscular Mycorrhizae (AM). A pot experiment with twenty-four treatment combinations was setup in the net house. The treatment combination consisted of 4 levels of P: P_0 - 0, P_{50} - 237.5 kg ha⁻¹ SSP, P_{75} - 355.5 kg ha⁻¹ SSP and P_{100} - 475 kg ha⁻¹ SSP, 3 levels of Zn: Zn₅₀- 5 Kg ha⁻¹ ZnSO₄, Zn₇₅-7.5 Kg ha⁻¹ ZnSO₄, Zn₁₀₀- 10 Kg ha⁻¹ ZnSO₄ and 2 levels of AM: I_0 -0 and I_{15} -15 g per pot. The main objective of the study was to reduce the antagonistic interaction between P and Zn by using AM fungi. The results of combined effects of application of P, Zn and AM revealed that P, Zn addition along with AM fungi improved plant growth parameters, plant nutrient content and total nutrient uptake (both above ground and underground portion) of the plant. The results indicated increase in plant height, root length and total nutrient uptake by increasing the level of P, Zn and Arbuscular Mycorrhizae. Also, Arbuscular Mycorrhizae enhanced plant growth by reducing Phosphorus or Zinc deficiency. Antagonistic effects of P and Zn addition on plant nutrient content and total nutrient uptake were absent due to

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application of AM fungi. Our results indicated that by using appropriate levels of AM fungi along with P and Zn, antagonistic interactions can be reduced for maximizing the plant nutrient content and nutrient uptake which may lead to improvement in plant growth and nutrition.

Keywords: Antagonistic; arbuscular mycorrhizae; nutrient interaction; phosphorus-zinc.

1. INTRODUCTION

Capsicum fruits are three to four lobbed, nonpungent which are eaten as raw, or cooked vegetables. Also, it is used in preparation of soups and, in stuffing and for salads or table purposes [1]. In plants, Phosphorus-Zinc interactions have been widely investigated [2]. The antagonistic P-Zn interaction further categorized according to whether high supplies of P application decreases, or does not decrease Zn concentration in plant [3]. Large amount of Phosphorus application causes a reduction in the Zn availability in plant tissue and grains [4]. High phosphorus and low Zinc supply in soil, enhances P concentration in plant tissue, which further leads to toxicity of Phosphorus [5]. By the application of high level of Phosphorus, there can be reduction in Zn translocation from roots to shoots. High Zn supply may entrap Zinc in roots by forming Zn-phytate [6]. Accumulation of Zn phosphates in roots may lead to uneven distribution of Zinc in the upper part of plant [4]. Mycorrhizae however enhance the plant nutrient status [7]. VAM (Vesicular-Arbuscular Mycorrhizae) plants have also improved plant nutrition [8] and increase plant tolerance. The main objective of this study was to evaluate the effect of AM fungi on the antagonistic interaction between P and Zn in Capsicum.

2. MATERIALS AND METHODS

The experimental area was situated at the net house of the Department of Soil Science and Water Management, Dr. Y S Parmar University of Horticulture and Forestry, Nauni, Solan, Himachal Pradesh, India. The pot experiment of present investigation was undertaken to evaluate the growth response of capsicum to varying levels of Phosphorus, Zinc and Arbuscular Mycorrhizae during the year 2017. The climate of experimental area falls in sub-temperate zone. The experiment was conducted in Complete Block Randomized Design. There were 24 treatment combinations comprised of 4 levels of Phosphorus (equivalent to 0, 50, 75 and 100% Recommended dose of Phosphorus) i.e. P_0 , P_{50} , P_{75} and P_{100} ; 3 levels of Zinc (equivalent to 50, 75 and 100% Recommended dose of $ZnSO₄$) i.e. $Zn₅₀$, $Zn₇₅$ and $Zn₁₀₀$; and 2 levels of Mycorrhizal Inoculation I_0 and I_{15} (0 and 15 g per pot) were replicated three times in CRD factorial design. By calculating dozes per 10 Kg of soil in pots, given treatments were applied. Nitrogen and Potassium application along with FYM were applied at the recommended rates.

2.1 Plant Analysis and Soil Analysis

Plant analysis was carried out by estimating plant growth parameters like root length and height of plant in centimeters. Plant samples (Root, fruit and shoot) were collected, washed, air dried, grounded and stored in paper bags for chemical analysis. For estimating nitrogen, 1 g of plant sample was digested in concentrated H_2SO_4 , in the presence of digestion mixture K_2SO_4 , CuSO₄ and Se powder (10:1:0.1).

Treatment combinations without AM	Treatment combinations with AM
Treatment ₁ : $P_0Zn_{50}I_0$	Treatment ₁₃ : $P_0Zn_{50}I_{15}$
Treatment ₂ : $P_0 Z n_{75} I_0$	Treatment ₁₄ : $P_0Zn_{75}I_{15}$
Treatment ₃ : $P_0 Zn_{100}I_0$	Treatment ₁₅ : $P_0Zn_{100}l_{15}$
Treatment ₄ : $P_{50}Zn_{50}I_0$	Treatment ₁₆ : $P_{50}Zn_{50}I_{15}$
Treatment ₅ : $P_{50}Zn_{75}I_0$	Treatment ₁₇ : $P_{50}Zn_{75}I_{15}$
Treatment ₆ : $P_{50}Zn_{100}I_0$	Treatment ₁₈ : $P_{50}Zn_{100}I_{15}$
Treatment _z : $P_{75}Zn_{50}I_0$	Treatment ₁₉ : $P_{75}Zn_{50}I_{15}$
Treatment ₈ : $P_{75}Zn_{75}I_0$	Treatment ₂₀ : $P_{75}Zn_{75}I_{15}$
Treatment ₉ : $P_{75}Zn_{100}I_0$	Treatment ₂₁ : $P_{75}Zn_{100}I_{15}$
Treatment ₁₀ : $P_{100}Zn_{50}I_0$	Treatment ₂₂ : $P_{100}Zn_{50}I_{15}$
Treatment ₁₁ : $P_{100}Zn_{75}I_0$	Treatment ₂₃ : $P_{100}Zn_{75}I_{15}$
Treatment ₁₂ : $P_{100}Zn_{100}I_0$	Treatment ₂₄ : $P_{100}Zn_{100}I_{15}$

Table 2. Different treatment combinations with or without AM

After digestion is completed, Nitrogen content of plant was determined by micro-kjeldahl method [9]. For estimation of other macro and micro nutrients, 1 g of plant samples including fruit, shoot and root samples were digested in di-acid mixture of $HNO₃$ and $HClO₄$ (4:1). Phosphorus in the extract was estimated by Vanado-molybdo yellow colour method, while Potassium was determined by flame photometric method. The Fe, Mn, Zn and Cu content of plant were determined in Atomic Absorption Spectrophotometer. For soil analysis, soil samples from each pot were collected for chemical analysis. Soil physicochemical properties like Organic carbon was estimated by using Walkley and Black Wet oxidation method [10]; available nitrogen estimated by Alkaline Potassium permanganate method [11]; available Phosphorus by stannous chloride reduced ammonium molybdate method using Olsen's extractant [12] and available Potassium was extracted by Ammonium Acetate method [13] and estimated on flame photometer. The micronutrient cations (DTPA extractable Fe, Mn, Zn and Cu) were estimated on atomic absorption spectrophotometer [14].

3. RESULTS AND DISCUSSION

3.1 Over Ground Parameters

The capsicum plant responded positively to varying levels of Phosphorus, Zinc and Mycorrhiza and with increase in levels of P, Zn and AM fungi, plant height increases with maximum plant height (67.3 cm) was obtained in $P_{100}Zn_{100}I_{15}$ whereas minimum plant height (49.8) cm) was observed in $P_0Zn_{75}I_0$ (Table 3). The results agree with observation of previous researchers, who observed that by using commercial microbial inoculants, growth and development of capsicum plant was affected [15]. The maximum total nitrogen content (9.91%) was observed in $P_{100}Zn_{100}I_{15}$ and minimum (7.32%) observed in $P_0Zn_{50}I_0$. These observations are well supported by those observed in literature showing that by the application of AM Fungi, there was high accumulation of Nitrogen in shoots of red pepper plant as compared to uninoculated controls [16]. The maximum total Phosphorus content (1.19%) was observed in $P_{100}Zn_{50}I_{15}$ and minimum $(0.67%)$ observed in $P_0Zn_{100}I_0$. Similar results were also reported by Tanwar and Aggarwal [17], that dual or multi inoculation of capsicum plants with *Glomus mosseae* had increased the Phosphorus content. Some studies also reported that application of Phosphorus increases P level in leaves [18]. The maximum total Potassium content (11.55%) was observed in $P_{100}Zn_{50}I_{15}$ and minimum (9.57%) observed in $P_{100}Zn_{100}I_0$. These results are in line with results showing that AM mycelium increases the potassium content in plants [19]. The maximum Fe content (500.14 mg kg^{-1}) was observed in $P_0Zn_{50}I_{15}$ and minimum $(273.00 \text{ mg kg}^{-1})$ in $P_{100}Zn_{100}I_0$. Halder and Mandal [20] reported similar results that Phosphorus application decreased the Fe concentration in shoots. The maximum total Mn content and Zn content (260.06 and 92.40 mg kg ¹) was observed in $P_0Zn_{100}I_{15}$ and minimum Mn and Zn contents (212.00 and 62.25 mg kg^{-1}) in $P_{100}Zn_{50}I_0$. Halder and Madal [20] in their studies observed that high P supply caused a decrease in concentration of manganese in shoots. These results are in accordance with previous studies reported that plants inoculated with some *Glomus* species had increased Zn content as compared to control [21]. The maximum total Cu content (53.45 mg kg^{-1}) was observed in $P_0Zn_{50}I_{15}$ and lowest (34.66 mg kg⁻¹) in $P_{75}Zn_{100}I_{15}$. Similar results were also reported in literature that by the application of phosphorus, there was decrease in Cu content in shoots of

pepper plant [22]. However, in three factor interaction, highest total nutrient uptake by over ground parameters (5.69 g plant⁻¹) was observed in $P_{100}Zn_{100}l_{15}$ and lowest (2.91 g plant⁻¹) was observed in $P_0Zn_{50}I_0$. These results are in line with previous literature that the increase in the uptake of essential nutrients was due to increased volume of soil under the inoculation by mycorrhizae [23]. The increase in plant nutrient content in above ground portion may be due to potentiality of mycorrhizal inoculated plants to take up nutrients through long thread-like structures (mycelium) from the soil which further helps in absorption of nutrients. Along with this, Arbuscular mycorrhizae also excrete some hormones, enzymes in root zone of plant, and change insoluble form of nutrients into soluble form, thus increase nutrient availability to the plant for absorption. This was also reported in literature that with the application of AMF, there is increase in nutrient content of major nutrients like N, P and K and micronutrients like Fe, Mn, Mg, Mo and Co [24]. The nutrient content of low mobility elements such as Phosphorus, Zinc and Copper may be increased by the application of AM fungi [25]. Also, it was reported that hyphal mycelium structures of mycorrhizae serve as a bridge between the soil and the root, promotes uptake of immobile nutrients by the plant [26].

3.2 Underground Parameters

In three-way interaction, among underground parameters, the maximum root length (13.0 cm) was observed in $P_{100}Zn_{100}I_{15}$. These results are in consonance with the findings reported that there was increase in root length and root weight of red pepper plants by the inoculation of *Methylobacterium oryzae* [16]. The maximum total Nitrogen and Phosphorus content (4.40 and 0.66%) was observed in $P_{100}Zn_{100}I_{15}$ and lowest Nitrogen and Phosphorus content (3.67 and 0.26%) in $P_0Zn_{50}I_0$. These results are similar with the results showing that the addition of AM fungi resulted in significantly higher Nitrogen (N) and Phosphorus content in the roots of red pepper plants compared to uninoculated controls [16]. The maximum total K content of 3.94% was observed in $P_0Zn_{100}I_{15}$ and minimum content (2.96%) in $P_{100}Zn_{50}I_0$. Similar results are in accordance with the results showing that mycorrhizal inoculation influences the Potassium content in plants [19]. The maximum total Fe and Cu content (141.20 and 30.69 mg kg^{-1}) was observed in $P_0Zn_{50}I_{15}$ and minimum content $(110.30$ and 11.59 mg kg⁻¹) in P₁₀₀Zn₁₀₀l₀. These results are in line with results reported by Halder

and Mandal [20] that application of phosphorus caused a decrease in the concentration of iron and copper in roots. Similarly, significantly higher total Mn content (142.00 mg kg^{-1}) was observed in $P_0Zn_{100}I_{15}$ and minimum content (126.05 mg kg^{-1}) in $P_{100}Zn_{50}I_0$. The decrease in Mn content in roots of capsicum plants with increasing the levels of Phosphorus and Zinc was also reported in literature [27]. The overall three-way interaction reveals that maximum total Zn content (46.38 mg kg⁻¹) was observed in $P_{100}Zn_{100}l_{15}$ and lowest total \overline{z} n content (33.03 mg kg $^{-1}$) was observed in $P_0Zn_{50}I_0$ (T₁) respectively. These results are similar to those studies showed that high level of soil available P or with high phosphorus application, zinc in roots may accumulate and metabolic disorder at cellular level may occur due to Phosphorus and zinc imbalance [28,12]. The higher total nutrient uptake by roots $(1.43 \text{ g plant}^{-1})$ was observed in $P_{100}Zn_{100}l_{15}$ and lowest (0.56 g plant⁻¹) was observed in $P_0Zn_{50}I_0$. These results are in accordance with the results reported that the mycorrhizal inoculation in pepper plant augmented plant growth by enhancing nutrient uptake through enlarging area of absorption in root, mobilization of available nutrients or by excretion of chelating compounds [29].

3.3 Soil Parameters

As given in Table 5, the maximum OC (1.47%) was observed. in $P_{75}Zn_{100}I_{15}$ and minimum OC (1.14%) was observed in P₀Zn₅₀I₀ which is statistically at par with $P_{50}Zn_{50}I_0$. The interaction reveals an increase in OC contents under mycorrhizal inoculated soils as compared to uninoculated soils. These findings are in accordance with the studies showing that AM fungus inoculated (+Myc) soil had significantly higher organic carbon than Uninoculated (−Myc) soil [11]. Among macronutrient content in soil, the maximum available Nitrogen and Potassium content in soil (213.00 and 131.00 mg kg^{-1}) were observed in $P_{75}Zn_{100}I_{15}$ and minimum available Nitrogen and Potassium content in soil (157.50 and 99.10 mg kg^{-1}) were observed in $P_0Zn_{50}l_0$. The maximum available phosphorus content in soil (56.60 mg kg⁻¹) was observed in $\mathsf{P}_{100}\mathsf{Zn}_{100}\mathsf{I}_{15}$ and minimum available phosphorus content in soil (34.27 mg kg^{-1}) was observed in $P_0Zn_{50}I_0$. The results are in line with the previous findings reported that, a combination of mycorrhizae and phosphate can increase available P on growing media [30]. It was also observed that with increasing levels of P application upto 100% recommended dose,

Treatments combinations	Plant height	N (%)	P $(\%)$	ĸ $(\%)$	Fe	Mn (mg/kg)	Zn (mg/kg)	Cu	Total uptake
	(cm)				(mg/kg)			(mg/kg)	(g/plant)
$P_0Zn_{50}I_0$	52.1	7.32	0.76	10.12	397.45	237.32	74.31	51.75	2.91
$P_0 Z n_{75} I_0$	49.8	7.48	0.74	10.25	372.00	244.75	83.66	49.28	3.11
$P_0 Zn_{100}I_0$	51.0	8.24	0.67	10.38	364.00	248.25	89.46	42.76	3.41
$P_{50}Zn_{50}I_0$	51.5	8.01	0.83	9.99	366.43	230.12	68.41	51.15	3.27
$P_{50}Zn_{75}I_{0}$	56.7	8.13	0.79	10.16	341.00	239.42	76.51	43.15	3.32
$P_{50}Zn_{100}I_0$	57.0	8.47	0.76	10.21	311.20	237.45	81.61	40.45	3.50
$P_{75}Zn_{50}I_{0}$	52.0	8.33	0.88	9.70	356.00	217.97	65.61	37.77	3.41
$P_{75}Zn_{75}I_0$	56.3	8.49	0.79	10.08	337.00	227.37	67.90	36.66	3.51
$P_{75}Zn_{100}I_0$	56.3	8.61	0.80	10.11	277.00	230.12	79.05	36.16	3.56
$P_{100}Zn_{50}I_0$	53.7	8.55	0.94	9.57	302.48	212.00	62.25	36.86	3.43
$P_{100}Zn_{75}I_{0}$	55.4	8.63	0.90	9.84	296.03	218.15	70.81	35.46	3.65
$P_{100}Zn_{100}I_0$	57.3	8.69	0.90	9.94	273.00	220.97	74.31	35.14	3.72
$P_0Zn_{50}I_{15}$	58.8	8.76	0.96	11.36	500.14	241.92	86.35	53.45	4.13
$P_0Zn_{75}I_{15}$	59.8	9.38	0.89	11.42	490.00	250.12	90.71	51.95	4.50
$P_0 Zn_{100}I_{15}$	61.0	9.60	0.86	11.55	429.00	260.06	92.40	43.75	4.63
$P_{50}Zn_{50}I_{15}$	61.5	9.11	1.02	11.21	428.50	233.02	76.19	51.95	4.31
$P_{50}Zn_{75}I_{15}$	63.3	9.46	0.95	11.24	422.50	251.32	84.75	47.19	4.92
$P_{50}Zn_{100}I_{15}$	63.7	9.62	0.91	11.27	399.00	257.92	87.24	43.16	4.99
$P_{75}Zn_{50}I_{15}$	62.0	9.45	1.15	10.42	399.50	228.07	72.15	50.46	4.59
$P_{75}Zn_{75}I_{15}$	62.9	9.61	1.09	11.12	391.00	233.77	80.69	42.15	5.28
$P_{75}Zn_{100}I_{15}$	63.0	9.76	0.96	11.22	386.00	245.92	84.16	34.66	5.51
$P_{100}Zn_{50}I_{15}$	63.7	9.57	1.19	10.24	384.00	221.62	68.31	40.25	4.76
$P_{100}Zn_{75}I_{15}$	65.4	9.69	1.12	11.07	367.00	229.77	74.56	39.61	5.46
$P_{100}Zn_{100}I_{15}$	67.3	9.91	1.07	11.18	356.00	235.57	77.23	38.91	5.69
Mean	58.4	8.87	0.91	10.57	372.76	235.54	77.86	43.09	4.15
$CD_{0.05}$	ΝS	0.06	0.04	0.19	10.10	4.01	1.90	0.97	0.11

Table 3. Growth response of varying levels of phosphorus, zinc and arbuscular mycorrhizae on over ground parameters

Table 4. Growth response of varying levels of phosphorus, zinc and arbuscular mycorrhizae on underground parameters

the NPK content of the soil (macronutrient content) increased along with the application of Zn as well as AM fungi, which may result due to increased availability of nutrients and may be ascribed to better soil health conditions. Among micronutrient content in soil, the maximum DTPA-Fe, Mn, Zn and Cu in soil (6.78, 2.35, 3.50 and 1.35 mg kg⁻¹) was observed in $P_{75}Zn_{100}I_{15}$ and minimum DTPA-Fe (5.29, 1.68, 0.72 mg kg-¹) was observed in $P_0Zn_0I_0$ whereas minimum DTPA-Zn was observed in $P_0Zn_{75}I_0$. The micronutrient content showed a fluctuating distribution pattern.

4. CONCLUSION

The main conclusion drawn from the present investigation is that plant parameters including

plant growth, nutrient content and nutrient uptake increased with increasing P and Zn application which were further enhanced by mycorrhizal application. At minimum level of application, antagonistic interaction between P and Zn affect Zn level in over ground parameters, which can be controlled by higher Zn application and mycorrhizal infection. However, in underground portion (root), there is formation of complexes by the higher application of P and Zn which leads to accumulation of Zn in the cells. The mycorrhizal inoculation not only improves the nutrient utilization but also control the effects of P and Zn antagonistic interactions. Though, soil parameters were affected a little but improved with inoculation and recommended application rates as well which finally leads to improvement in soil health.

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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