

Fortification of Wheat (*Triticum aestivum* L.) with Zinc and Manganese

Amandeep Kaur¹, Simerpreet Kaur²* Didar Singh³, Satnam Singh⁴ and Manpreet Singh⁵

* PG Department of Agriculture, Khalsa College, Amritsar-143 005 (Punjab)

ABSTRACT

The field experiment on fortification of wheat (Triticum aestivum L.) with zinc and manganese was conducted during the Rabi season of 2018 at Students' Research Farm, Department of Agriculture, Amritsar. The experiment was laid out in randomized block design with twelve treatments with four replications viz., T1: Control; T2: ZnSO4 @ 30 kg/ha in wheat at sowing time; T3: ZnSO4 @ 60 kg/ha in wheat at sowing time; T4: ZnSO4 @ 90 kg/ha in wheat at sowing time; T5: MnSO4 @ 0.5% solution spray 2-4 days*; T6: MnSO4 @ 1.0% solution spray 2-4 days*; T7: ZnSO4 @ 30 kg/ha in wheat at sowing + MnSO4 @ 0.5% solution spray 2-4 days*; T8: ZnSO4 @ 60 kg/ha in wheat at sowing + MnSO4 @ 1.0% solution spray 2-4 days*, T9: ZnSO4 @ 90 kg/ha in wheat at sowing + MnSO4 @ 0.5% solution spray 2-4 days*, T10: ZnSO4 @ 30 kg/ha in wheat at sowing + MnSO4 @ 1.0% solution spray 2-4 days*, T11: ZnSO4 @ 60 kg/ha in wheat at sowing + MnSO4 @ 0.5% solution spray 2-4 days*, T12: ZnSO4 @ 90 kg/ha in wheat at sowing + MnSO4 (a) 1.0 % solution spray 2-4 days. The soil of experimental site was sandy loam in texture, medium in organic carbon (0.40%) with 194.62, 19.75 and 248.68 kg/ha of available N, P and K, respectively along with 0.60 and 3.81 ppm of available Zn and Mn, respectively. Results revealed that T12 recorded significantly highest growth parameters and yield attributing characteristics as compared to other treatments except T11 and T4 which were at par with T12. The soil parameters such as pH (7.41), CaCO₃ (2.85%), available P (16.37 kg/ha) decreased in T12 whereas values of EC (0.34 dS/m), OC (0.44%) increased in the same treatment after harvesting of the crop. The highest grain and straw uptake of Zn (155.57 and 153.38 g/ha) and Mn (89.36 and 80.47 g/ha) were observed significantly higher in T12 than other treatments except T11, T4 for Zn and T11, T6 for Mn, respectively.

Key Words: Fortification, micronutrients, nutrient uptake, soil and foliar application, wheat.

INTRODUCTION

Wheat (*Triticum aestivum* L.) native of South West Asia, is one of the most important staple and winter cereal crop that has been labeled as king of cereals (Akhtar *et al*, 2018). India is the 2^{nd} largest wheat producer country in the world occupying about 29.58 mha of area alone producing 99.70 Mt (Anon, 2018). Due to poor quality food intake about 50 per cent of the world's population is suffering from micronutrient malnutrition (Aziz *et al*, 2019). Health index in developing countries is declining due to low levels of malnutrition in the diet (Jankowska *et al*, 2012). About 75 percent of

world's population suffers from inadequate intake of micronutrients in diet as Zn, problems of anaemia, weekened immune system contributing 40 per cent of women and children all over world. (Sangeetha and Premakumari, 2010).

Zinc (Zn) is considered as one of the prime and essential micronutrient for growth of plant which is absorbed by plant roots in the form of Zn^{2+} . It has role in plant metabolism and its deficiency has effects on plant growth though there are number of different mechanisms by which plants can tolerate Zn deficiency via Zn translocation in both shoot and root of wheat plant. Zinc is essential

Corresponding Author's Email: sehgalenv@gmail.com

for activating plant's enzymatic systems, protein synthesis (Hafeez *et al*, 2013), photosynthesis, reproduction of genetic material (DNA) during cell division and the synthesis of chlorophyll and carbohydrates (Kobraee *et al*, 2011). Zinc deficiency not only retards the growth and yield of plants but also affects human beings (Ayalew, 2016) with malnutrition, neuronal disorders of susceptibility to various infectious diseases (Hafeez *et al*, 2013).

Manganese (Mn) also plays an important role in plant metabolic processes as an essential micronutrient. It is available to plants as Mn²⁺ and can be easily transported into root cells then translocate to the shoots, where it is finally accumulated. Mn plays an important role being cofactor of enzymes like MnSOD (superoxide dismutase), MnCAT (catalase) and TCA (decarboxylases of tricarboxylic acid) (Millaleo et al, 2010). Mn deficiency is also one of the common occurring deficiencies in the world which may lead to poor reproductive performance, growth retardation, congenial malformation in human beings (Aschner and Erickson, 2017). In case of plants. Mn deficiency is highly sensitive and revealed noticeable decline in yield of crop as it affects photosynthesis by causing interveinal chlorosis (Jhanji et al, 2014).

Therefore, to reduce the deficiency of essential nutrients, agronomic strategies are used to increase micronutrient content and their bioavailability for human nutrition in the edible parts of crops by adding micronutrient fertilizers to the soil or plant leaves, called fortification (Kadam *et al*, 2017). Micronutrients such as Zn, Mn are considered as essential nutrients. Keeping the importance of nutrients in view, the study was undertaken to assess the effect of application of Zn and Mn at different rates on physico-chemical properties of soil including the status of Zn and Mn in soil, their effect on yield of wheat and on nutrient uptake in wheat grains.

MATERIALS AND METHODS

A field experiment was initiated during winter (*Rabi*) seasons of 2018-19 at Students' research

farm, Khalsa College, Amritsar, Punjab, India. The climate of the experimental site is semi-arid with dry hot summers and cold winters with average annual rainfall of 75 cm, 80 percent of which is received through south-west monsoon during July-September. Soils are sandy loam in texture. A uniformity trial on wheat was undertaken during Rabi 2018-19 to ensure uniform soil physico-chemical status in the entire field. The soil (0-15 cm layer) had pH 7.61 (1 : 2 soil and water ratio), organic carbon (0.40%), available N (194.48 kg/ha), P (19.78 kg/ha) and K (247.91 kg/ ha) to be observed before sowing. The soil contained diethylene triamine penta acetate (DTPA)-extractable Zn and Mn were 0.57 mg/kg and 3.78 mg/kg soil. The experiment was laid out in a randomized block design (RBD) and replicate four times. Unnat PBW 343 wheat variety was used in this study. Experiment comprises of T1: Control; T2: ZnSO4 @ 30 kg/ha in wheat at sowing time; T3: ZnSO4 @ 60 kg/ha in wheat at sowing time; T4: ZnSO4 @ 90 kg/ha in wheat at sowing time; T5: MnSO4 @ 0.5% solution spray 2-4 days*; T6: MnSO4 @ 1.0% solution spray 2-4 days*; T7: ZnSO4 @ 30 kg/ha in wheat at sowing + MnSO4 @ 0.5% solution spray 2-4 days*; T8: ZnSO4 (a) 60 kg/ha in wheat at sowing + MnSO4 @ 1.0% solution spray 2-4 days*, T9: ZnSO4 (\hat{a}) 90 kg/ha in wheat at sowing + MnSO4 (a) 0.5% solution spray 2-4 days*, T10: ZnSO4 (a) 30 kg/ha in wheat at sowing + MnSO4 (a) 1.0% solution spray 2-4 days*, T11: ZnSO4 @ 60 kg/ ha in wheat at sowing + MnSO4 (a) 0.5% solution spray 2-4 days*, T₁₂: ZnSO4 @ 90 kg/ha in wheat at sowing + MnSO4 (a) 1.0 % solution spray 2-4 days*).

N, P and K were applied through urea and di ammonium phosphate (DAP). Nitrogen was applied in three splits *i.e.*, one half at sowing and remaining dose was applied before first irrigation and 15 days later than that. Zn was applied as basal dose at time of sowing at 30, 60 and 90 kg/ha, respectively according to treatments whereas Mn was applied as foliar application @ 0.5% and 1.0% before 2-3 d of first irrigation and subsequent three sprays were applied at 10 d intervals. Wheat crop was raised by

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following the standard recommended package of practices and harvested in the second fortnight of April during the year. Macro and micro-nutrients content at the time of harvesting in the plant was determined by drying the samples in hot-air oven at $60^{\circ}C \pm 2^{\circ}C$ till a constant dry weight obtained. Nitrogen content in soil samples was analyzed by using Kjeldahl's apparatus and expressed as kg/ ha. The available P was determined by using Olsen method (Olsen et al, 1954). Also, available K was determined by Merwin and Peech (1950) using the flame photometer and expressed as kg/ha. Available Zn and Mn were also determined by DTPA extractable solution and analysed by using Microwave Plasmaatomic Emission Spectrometer (Agilent 4200 MP-AES). The micro-nutrients uptake was computed by multiplying the nutrient content with respective crop yield and was expressed as nutrient uptake in g/ha. The data obtained from study, were analysed statistically using CPCS-1 software developed by the department of Mathematics and Statistics, PAU,

Ludhiana. The comparison was made at 5 percent level of significance.

RESULTS AND DISCUSSION

Soil parameters : pH, EC, OC and CaCO,

The data pertaining to soil pH, EC, OC and $CaCO_3$ as influenced by application of different levels of Zn and Mn through different methods of application at harvest of wheat crop is presented in Table 1 which depicted that various treatments had shown no significant results on soil parameters.

Nutrient availability

The effect of different treatments on soil available N, P, K, Zn and Mn after harvesting of wheat is presented in Table 1 which represented that among various treatments, T_{12} (201.09 kg/ha) recorded the significant highest amount of nitrogen in soil after harvest followed by T_{11} (200.63 kg/ha) and T_4 (200.11 kg/ha) and proved significantly superior to rest of the treatments. It was due to the fact that Zn

Table 1. Effect of different treatments on soil pH, EC (dS/m), Organic Carbon (OC%) and CaCO₃ (%),available N (kg/ha), available P (kg/ha), available K (kg/ha), available Zn (mg/kg) and available Mn (mg/kg) of soil after harvest of wheat crop.

Treatment	pН	EC	OC	CaCO3	Available nutrients						
					Ν	Р	K	Zn	Mn		
T1	7.59	0.29	0.40	2.95	194.62	19.75	248.68	0.60	3.81		
T2	7.57	0.29	0.40	2.93	197.34	18.56	251.35	0.75	5.45		
T3	7.54	0.29	0.41	2.91	198.54	17.59	252.42	0.84	6.02		
T4	7.53	0.29	0.43	2.93	200.11	16.84	253.21	0.95	8.57		
T5	7.55	0.31	0.41	2.92	196.31	19.21	249.93	0.66	4.21		
T6	7.53	0.31	0.42	2.89	196.40	18.99	249.61	0.69	4.98		
Τ7	7.54	0.31	0.40	2.88	197.39	18.32	251.37	0.77	5.74		
Т8	7.51	0.31	0.41	2.87	197.99	18.25	251.83	0.80	5.89		
Т9	7.52	0.34	0.42	2.88	198.93	17.48	252.68	0.87	6.32		
T10	7.50	0.32	0.42	2.86	199.26	17.09	252.91	0.91	7.63		
T11	7.42	0.34	0.43	2.85	200.63	16.56	253.58	0.99	8.76		
T12	7.41	0.34	0.44	2.85	201.09	16.37	254.00	1.03	9.49		
CD (0.05%)	NS	NS	NS	NS	0.90	0.51	NS	0.04	0.75		

*before first irrigation and three sprays afterwards at 10 days intervals.

and N showed positive (synergistic) effect with each other which helped to improve the available N in soil after harvesting the crop. When Zn application was adequate, both soil and foliar N applications significantly increased grain Zn concentration. Nitrogen application remained ineffective on grain Zn when Zn supply was sub-optimal because of positive effect of Zn and N in improving the grain Zn concentration (Cakmak, 2010). Also, Zn exerts a great influence on basic plant life processes such as nitrogen metabolism-uptake of nitrogen (Zeidan et al, 2010). Also, Mn along with Zn has an effect on protein biosynthesis by adjusting the activity of peptidases and controlling protein metabolism which is due to the availability of nitrogen in higher amount. Therefore, application of Zn and Mn showed highest available N content (Stepian and Wojtkowiak, 2016). P showed negative effect with Zn and Mn which results in the reduction of available P with increasing levels of zinc and manganese application either alone or in combination with each other. It was due to the fact that P is present in unavailable form in soil (Kanubhai, 2013). In case of low availability of Zn and Mn also enhanced expression of Ptransported genes in root cells and P accumulation in increased in plants. Also, soil or foliar application of micronutrient fertilizers reduced shoot and grain P concentrations. The highest available K was found in T₁₂ attributed to direct K addition in the potassium pool of the soil. Also, it might be due to reason that K have positive relation with Zn and Mn both (Adekiya et al, 2018). The experimental soil being low in available Zn might have resulted in increased available Zn linearly with the increasing level of Zn application. It could possibly caused solubilization of native Zn with increase in the rate of Zn application. Maximum available manganese (9.49 mg/kg) in T₁₂ was observed because Mn availability to plants is controlled by its concentration in soil solution, which depends on the chemistry of soil matrix and Mn forms (Rehman et al, 2016). Mn in soil solution generally represents a very small proportion of total soil Mn, exists in equilibrium with mineral forms and with organically complexed and exchangeable Mn.

However, supplementation may increase the uptake and availability to plants.

Effect of Different Levels of Zn and Mn Application on Growth and Yield Attributes of Wheat Crop

Periodic plant height (cm)

Plant height is an important index of the plant development. It gives an idea to predict the growth rate and yield of crop. The data (Table 2) indicated that plant height was significantly increased in T_{12} as compared to T_1 due to the reason that Zn and Mn have structural role in chlorophyll. These elements can be easily sprayed on leaf, thus leaves chlorophyll concentration increased by micronutrient soil and foliar application, which in turn, lead to an increase in plant height and yield. Also, Zn increased plant height via increasing internodes distances (Bameri *et al*, 2012).

Leaf area index (LAI)

Leaf area index is an important plant growth indices that determines the capacity of plants in trapping solar energy for photosynthesis and has marked influence on growth and yield of crop. The greater leaf area index in T_{12} at 120 DAS could be attributed to significant increases in leaf expansion i.e. length and breadth due to Zn application. Greater leaf expansion was ascribed to higher rate of cell division and cell enlargement in wheat plant.

Dry matter accumulation (DMA)

DMA is most important parameter and has a marked effect on final yield realization of crop. The optimum accumulation of dry matter followed by adequate partitioning of assimilates to the developing sink and enables the crop to attain its true yield potential. Dry matter accumulation increased in all the treatments with the increase in age of crop.

Number of tillers per m²

Among the yield contributing characters, the number of tillers per square meter not only reflects the proper crop establishment but has a great effect

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on grain yield because it contribute to higher number of grains to increase the yield of wheat crop. The important role of Zn in initiation of primordial for reproductive parts and bio-synthesis of IAA might had resulted in better development of these yield contributing characters (Sharma *et al*, 2017).

Number of grains per ear and test weight

Data on number of grains per ear and test weight (Table 2) showed non- significant increase in both parameters in T_{12} than rest of the treatments. The number of grains per panicle is an important factor influencing the final grain yield of wheat and depends on the genetic makeup of genotypes (Nadim *et al*, 2012).

Grain yield (q/ha)

The data with respect to grain yield highlighted that T_{12} produced significantly higher yield as

compared to other treatments except T_{11} which is at par to T_{12} (Table 2). It was due to the favorable effect of Zn and Mn on productivity of wheat and might be due to function of Zn as catalyst or stimulant in most of the physiological and metabolic processes and metal activator of enzymes helping in carbohydrate and protein synthesis. While Mn is absolutely required for photosynthesis which cannot be carried out without this element because Mn is the central part of the oxygen evolving complex at photosystem II. The increase in yield attributes may be due to increased supply of available Zn and Mn to plants by way of its addition to soil which resulted in proper growth and development (Ullah, *et al*, 2017).

Straw yield is an important parameter of biological yield to evaluate its productivity index for judging the ultimate performance of wheat

Treatment	Plant height at harvest	ai	DMA at harvest	Tillers per m ²	Grains per ear	Test weight	Grain yield	Straw yield	Biological yield
		120 DAS							
T1	91.60	2.72	99.5	190.08	41.34	37.80	42.35	55.56	97.91
T2	92.35	2.89	101.82	204.78	41.41	37.81	44.57	57.89	102.46
T3	93.76	2.97	103.76	219.38	41.46	37.92	44.83	58.39	103.22
T4	94.63	3.07	106.28	235.69	41.52	38.02	45.57	59.07	104.64
T5	93.62	2.98	104.01	221.42	41.48	37.96	44.98	58.74	103.72
T6	94.24	3.08	106.42	240.78	41.55	38.04	45.79	59.59	105.38
T7	96.04	3.11	108.63	256.29	41.64	38.32	46.68	60.87	107.55
T8	96.31	3.17	110.59	259.44	41.70	38.35	46.79	60.95	107.74
Т9	98.07	3.19	111.84	266.83	41.72	38.40	47.03	61.81	108.84
T10	98.76	3.22	113.68	270.15	41.79	38.43	47.45	61.99	109.44
T11	100.55	3.23	114.02	284.36	41.81	38.48	48.25	62.36	110.61
T12	102.02	3.26	114.28	289.00	41.90	38.67	48.54	62.71	111.25
CD	1.76	0.25	2.20	2.16	NS	NS	0.78	0.70	1.02
(0.05%)									

Table 2. Effect of different treatments on plant height (cm), leaf area index, dry matter accumulation (kg/ha) and number of tillers per m², grains per ear, test weight (g), grain yield (q/ha), straw yield (q/ha) and biological yield (q/ha) at harvesting of wheat.

* before first irrigation and three sprays afterwards at 10 days interval

crop and it has economic value as it is fed to the cattle. Biological yield refers to the total dry matter accumulation of the plant system. The combined effect of (soil + foliar application) micronutrients could be due to possibility of exploring greater soil volume for nutrient absorption, which might have resulted in increasing straw yield and biomass of crop (Kanubhai, 2013). Highest biological yield in T_{12} was due to the fact that soil + foliar application of Zn and Mn, respectively played a significant role in the crop growth, involving in photosynthesis processes, respiration and other biochemical and physiological activates and thus importance in improving higher yields (Zeidan *et al*, 2010).

Grain and straw Zn and Mn uptake (g/ha)

It was clearly evident from the data in Table 3 that when wheat crop was fertilized with Zn might have improved the nutritional environment of rhizosphere, which resulted in greater nutrient uptake by the crop and caused higher metabolic and photosynthetic activity in plant leading to the higher yield. There was significant difference in Zn uptake by wheat straw due to different micronutrient application through soil either alone or in combination with foliar Mn application. It was attributed to the fact that with the increasing level of the Zn, the straw Zn uptake also increased. However, the increasing dose of Zn showed a little increase in the uptake of Mn because wheat fertilized

with Zn improved the nutritional environment of rhizosphere, which resulted in greater uptake of zinc by crop and this caused higher metabolic and photosynthetic activity in plant leading to higher plant (Abbas *et al*, 2009).

The highest grain Mn uptake i.e. 89.36 g/ha was also noticed in treatment T₁₂ which was significantly higher than that of all other treatments except T_{11} due to higher absorption of Mn in foliage of wheat but its further translocation to grain was not in same proportion, even though an increase in Mn uptake by grain with increase in rate of application of MnSO₄ was noticed (Kanubhai, 2013). The Zn uptake was important in enhancing yield and nutrient content. Considerable increase in either nutrient content or in yield may increase uptake. The uptake of Zn is a function of its improved metabolic reactions, activation of enzymes that leads to improvement in quality parameters (Jeet et al, 2012). The significant difference in straw Mn uptake of wheat may be due to the fact that the continuous uptake of Mn by roots takes place through xylem tissue of plant. The significant positive correlation of Mn influx with grain Mn uptake supported the higher direct Mn supply to grain, hence produced higher Mn uptake in straw of wheat (Jhanji et al, 2014). Also, the increase in Mn solubility by microbial or chemical mobilization would increase Mn uptake by wheat plant (Abbas et al, 2009).

Treatment	Zn uptake		Mn uptake		Treatments	Zn uptake		Mn uptake	
	Grain	Straw	Grain	Straw		Grain	Straw	Grain	Straw
T1	86.43	86.11	44.72	42.39	Τ7	135.46	134.46	65.51	56.57
T2	102.95	94.01	59.45	52.90	Τ8	138.87	137.5	67.95	58.30
T3	116.78	101.36	69.95	59.22	Т9	143.39	144.14	76.70	64.33
T4	128.59	109.22	79.98	70.18	T10	147.14	147.59	79.90	66.47
T5	117.44	99.97	72.60	69.94	T11	153.77	150.72	84.99	79.56
T6	128.12	107.85	77.58	72.02	T12	155.57	153.38	89.36	80.47
				~	CD (p=0.05)	8.46	9.45	4.52	1.20

Table 3. Effect of different treatments on Zn and Mn uptake (g/ha) in grain and straw of wheat crop.

* before first irrigation and three sprays afterwards at 10 days interval

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CONCLUSION

The present research revealed that micronutrients and their application methods had significant effect on the growth and yield of wheat. Based on the results obtained, it might be concluded that combined application of micronutrients could be useful for improving the nutrient status, physiological performance of wheat plants. It proved best for fortification to enhance their content in grains. It is pertinent to say that increase in the contents of zinc and manganese in wheat grains is possible by applying them with different combinations to have Zn and Mn enriched wheat grains.

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