Journal of Pharmacognosy and Phytochemistry

Available online at www.phytojournal.com

E-ISSN: 2278-4136
P-ISSN: 2349-8234
JPP 2019; SP4: 131-134

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(Special Issue- 4)
National Seminar
“Role of Biological Sciences in Organic Farming”
(March 20, 2019)

Fortification of maize (*Zea mays* L.) by methods and time of Zinc application

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Abstract
The field experiment was conducted at Khalsa College, Amritsar to evaluate the fortification of maize, (*Zea mays* L.) by methods and time of Zn application during *kharif* 2016 on sandy loam soil. Zn as in soil (25 kg ha^{-1}) or foliar application (1.0% Zn foliar spray at different stages) alone and in combination (25 kg ha^{-1} in soil and 0.5% Zn as foliar spray were evaluated in randomized block design having three replications. Results showed that combined application of Zn in soil and foliar spray (0.5%) significantly improved plant height, leaf area index, dry matter accumulation, cob length, no of grains/cob, grain yield and biological yield. Similarly, with combined application Zn in soil and foliar spray produced significantly more Zn content (ppm) in grain. Zn foliar application at lateral stage produced higher grain yield, yield attributes and Zn content in grain of maize than earlier stages. These results suggested that combined application of Zn as soil and foliar spray can improve the performance of maize hybrids.

Keywords: Fortification, Zn, Maize.

Introduction
As maize is an exhaustive crop, it requires large amount of nutrients from soil during its vegetative growth. Maize is one of the most susceptible cereal crops to Zn deficiency. Zn applications are reported to increase maize grain yield around the world (Harris *et al*. 2007, Hossain *et al*. 2008). Zn deficiency is frequently equal in plant and human. Zn deficiency region in humans are reported to be Zn deficient soil region and are prevalent, in India, China, Pakistan and Turkey. Worldwide incidence of Zn deficiency in soil is becoming more important due to impact on human health (Singh *et al*. 2005).

Biofortification is a process in which plants are allowed to take up the minerals from the soils and immobilize them in the grains so as to produce nutritionally rich grains that support dietary requirement of humans. Moreover, the proper method of nutrient application can be another approach for better uptake and utilization of Zn. Amongst the different method, the foliar spray of micronutrients is efficient in enhancement of crop productivity (Savithri *et al*. 1999). Response to the foliar application depends upon several determining factors like penetration, absorption and translocation of nutrients. So there is need to determine the different schedules for foliar application according to the growth stages of maize. (*Zea mays* L.) Keeping in view the above scenario, a field experiment was planned in order to assess the growth response and yield potential of maize hybrid under varying Zn application methods.

Material and Methods
The present investigation entitled “Fortification of maize (*Zea mays* L.) by methods and time of Zn application” was conducted during the *kharif* of 2016 at the Students' Research Farm, Khalsa College, Amritsar. The soil of the experimental site was sandy loam in texture with normal soil pH and electrical conductivity, low in organic carbon, available N and Zn, high in available P and K. The experiment was laid out in randomized block design having three replications. In each replication there are fourteen treatments having methods of Zn application at different stages of maize. In T_1: control (recommended dose of NPK, with no Zn), T_2: 25 kg $ZnSo_4$ ha^{-1} as soil application, T3: foliar spray at knee stage ($ZnSo_4$ application @ 1% solution) T_4: foliar spray at tasseling stage ($ZnSo_4$ application @ 1% solution), T_5: foliar spray

at grain filling stage ($ZnSo_4$ application @ 1% solution), T6: foliar spray at knee+ tasseling stage ($ZnSo_4$ application @ 1% solution), T7 foliar spray at tasseling + grain filling stage ($ZnSo_4$ application @ 1% solution), T8 foliar application at knee+ tasseling+ grain filling stage ($ZnSo_4$ application @ 1% solution), T_9: application of (T_2 +T_3), T10: application of (T_2+ T_4), T_{11}: application of (T_2+T_5), T_{12}: application of (T_2+ T_6), T_{13}: application of (T_2+ T_7), T_{14}: application (T_2+T_8). The pre treated seeds of variety PMH-1 were sown on ridges by dibbling on 5th June 2016. Recommended nitrogen (Urea 125) applied 1/3 at sowing, 1/3 knee high and 1/3 pre tasselling stage. Zinc hyptahydrate (21 per cent) applied in soil @ 25 kg ha^{-1}at sowing in T2, T9, T10, T11, T12, T13, and T14. Zn was applied @ 1 per cent in T_3, T_4, T_5, T_6, T_7, T_8 treatments and 0.5 per cent in all the treatments in which Zn is applied through soil + foliar method i.e T_9, T_{10}, T_{11}, T_{12}, T_{13}, T_{14}. The maize crop was harvested manually with sickles on 07 sept, 2016. Seven days after harvesting, the cobs were dehusked manually and were allowed to dry for seven days and thereafter the threshing was done using maize dehusker cum thresher. Zn content in grains was calculated by Atomic absorption spectrophotometer after wet digestion of grains. The digestion process is as: weight 2.5 g of sample into a 600 ml beaker. Add 25 mL of concentrated HNO_3, cover with a watch glass, and boil gently for 30-45 minutes to oxidize all the easily oxidizable material. Cool the solution and slowly add 10 mL of 70% $HC1O_4$. Boil very gently until the solution is nearly colorless. Do not allow the solution to go to dryness. Statistical analysis of the data recorded was done as per RBD design using EDA software developed by the Department of Mathematics and Statistics, PAU, Ludhiana.

Results and Discussion

Growth

The data presented in Table 1 showed significant difference occurred for growth parameters viz. plant height, LAI, dry matter accumulation due to application of Zn through different methods at different time. Maximum growth parameters were recorded in T_{14} (25 kg/ha Zn in soil at sowing + foliar application at knee, tasseling and grain filling stage). All Zn foliar applications from T_3 to T_8 were at par with each other. Zn soil application (T_2) is significantly higher growth than control (T1) and T_3 to T_8. Vasconcelos *et al.* (2011) [7] also gave similar results. It was concluded that in foliar applications, the treatments in which foiliar spray was applied at knee stage gave good result followed by tasseling and grain filling in comparison to control treatment but it was statistically at par among each other. The same pattern was seen when foliar application was added with soil application, it show that Zn application at initial stages enhance growth in comparison to application at later stages. Zn foliar application in initial stages and Zn soil application give best result. The crop response to the late foliar application i.e. at grain filling was non- significant. Greater LAI could be attributed to significant increases in leaf expansion i.e. length and breadth due Zn application. Greater leaf expansion in maize was ascribed to higher rate of cell division and cell enlargement. Similar results were reported by Jaliya *et al.* (2008) [3].

Table 1: Effect of methods and time of Zn application on growth in maize (*Zea mays* L.)

Treatments		Plant height	LAI	DMA
Control (only recommended dose of NPK fertilizer)	T_1	183.59	3.6	111.86
Zn soil application (25kg/ha)	T_2	202.5	4.12	122.15
Zn Foliar at knee stage (1%)	T_3	185.36	3.69	114.78
Zn Foliar at tasseling (1%)	T_4	184.54	3.67	113.47
Zn Foliar at Grain filling (1%)	T_5	183.78	3.63	112.78
Zn Foliar at knee + tasseling (1%)	T_6	187.41	3.70	116.36
Zn Foliar at tasseling + Grain filling (1%)	T_7	184.72	3.68	115.47
Zn Foliar at knee + tasseling +. Grain filling (1%)	T_8	187.18	3.74	117.04
Zn_{soil} + Zn_{foliar} at knee stage (0.5%)	T_9	204.29	4.23	123.41
Zn_{soil} + Zn_{foliar} at tasseling (0.5%)	T_{10}	203.03	4.18	123.6
Zn_{soil} + Zn_{foliar} at Grain filling (0.5%)	T_{11}	202.66	4.12	122.82
Zn_{soil} + Zn_{foliar} at knee + tasseling (0.5%)	T_{12}	207.05	4.30	126.59
Zn_{soil} + Zn_{foliar} at tasseling +Grain filling (0.5%)	T_{13}	203.61	4.19	125.19
Zn_{soil} + Zn_{foliar} at knee + tasseling + Grain filling (0.5%)	T_{14}	209.02	4.30	128.55
CD (p = 0.05)		7.40	0.24	6.62

Yield attributing Characters

Number of cobs per plant has linear relation with yield. The data present in the Table 2 indicated that the Zn application showed non-significant difference in number of cobs per plant. Whereas different treatments showed significant effect on cob length and number of grains cob^{-1}.The maximum cob length (19.0 cm) and number of grain cob^{-1} (325) was observed in treatment T_{14} (25 kg ha^{-1} Zn in soil at sowing +3 foliar application at knee, tasseling and grain filling stage) and lowest cob length (17.18 cm) and number of grains (293.33) in control treatment T_1. It may be due to reason that growth parameters such as plant height, leaf area index etc were better when 25 kg ha^{-1} Zn in soil at sowing +3 foliar application at knee, tasseling and grain filling stage (T_{14}) is applied in comparison to zero Zn (T_1). All the treatments in which Zn is applied through soil + foliar method i.e T_9, T_{10}, T_{11}, T_{12}, T_{13}, T_{14} and Zn soil application (T_2) are significant over control treatment. All the only foliar applications i.e T_3, T_4, T_5, T_6, T_7, T_8 are at par with control treatment (T_1). The probable reason for lesser grain number cob^{-1} was Zn deficiency which reduced biomass production traits (i.e. leaf area and light capture of plant which could be primarily relate to number of grain. Aruna *et al.* (2006) [1] also found the similar results.

Test weight

Data of 100 grain weight presented in Table 2 showed non-significant result. Numerically highest test weight (20.6 gm) and minimum test weight (19.95 gm) was obtained from T_{14} and T_1 respectively. It might be due to Zn application and its effect on translocation of constitutes into grains. The result suggested that the Zn supply might have enhanced the source efficiency (more dry matter accumulation per unit area) as well kernel weight. The treatments in which Zn was applied through soil application show more test weight in comparison control and foliar applications. These results are in conformity with the finding of Hossain *et al.* (2011) [2]

Table 2: Effect of methods and time of Zn application on yield and yield attributing characters of maize in maize (*Zea mays* L.)

Treatments		Number of cobs per plant	Cob length (cm)	Grains cob^{-1}	Test weight (gm)	Grain yield (q ha^{-1})
Control (only recommended dose of NPK fertilizer)	T_1	1.06	17.18	293.33	19.95	47.48
Zn soil application (25kg/ha)	T_2	1.13	18.63	316	20.5	51.31
Zn Foliar at knee stage (1%)	T_3	1.06	17.37	295	20.05	48.72
Zn Foliar at tasseling (1%)	T_4	1.06	17.32	293	19.98	48.64
Zn Foliar at Grain filling (1%)	T_5	1.06	17.18	292	19.95	48.1
Zn Foliar at knee + tasseling (1%)	T_6	1.06	17.5	298	20.1	49.75
Zn Foliar at tasseling + Grain filling (1%)	T_7	1.06	17.39	295	20.08	49.48
Zn Foliar at knee + tasseling +.Grain filling (1%)	T_8	1.06	17.6	300	20.12	49.87
Zn_{soil} + Zn_{foliar} at knee stage (0.5%)	T_9	1.13	18.81	319	20.55	51.786
Zn_{soil} + Zn_{foliar} at tasseling (0.5%)	T_{10}	1.13	18.72	317	20.52	51.65
Zn_{soil} + Zn_{foliar} at Grain filling (0.5%)	T_{11}	1.13	18.64	316	20.5	51.45
Zn_{soil} + Zn_{foliar} at knee + tasseling (0.5%)	T_{12}	1.13	18.95	322	20.57	52.56
Zn_{soil} + Zn_{foliar} at tasseling +Grain filling (0.5%)	T_{13}	1.13	18.77	320	20.55	52.37
Zn_{soil} + Zn_{foliar} at knee + tasseling + Grain filling (0.5%)	T_{14}	1.13	19.02	325	20.6	52.78
CD (p = 0.05)		NS	1.26	10.06	NS	3.43

Grain yield

Grain yield is the final outcome of all the management and various factors influencing growth and yield attributing characters during the life cycle of the crop. The real efficiency of different treatments can be judged by their ultimate effect on economic yield of crop. The data with respect to grain yield presented in Table 2 showed that Zn application had significant effect on grain yield of maize. Highest grain yield (52.78 q ha^{-1}) was recorded when Zn @ 25 kg ha^{-1} applied in soil at sowing + foliar application at knee, tasseling and grain filling stage (T_{14}). While lower yield (47.48 q ha^{-1}) was recorded when recommended dose of N,P,K is applied but Zn application is absent i.e T_1 (control treatment). Data showed that grain yield of T_{14} increased 11.16% than control (T_1). Zn application in soil at sowing (T_2) give yield (51.31 q ha^{-1}) was significantly better from control treatment (T_1). All the treatments in which only foliar Zn application was applied i.e T_3, T_4, T_5, T_6, T_7, T_8 are at par with each other and also with control (T_1). All the Zn soil + foliar treatments i.e T_9, T_{10}, T_{11}, T_{12}, T_{13}, T_{14} and Zn soil application at sowing (T_2) were significantly batter from control treatment (T_1) but these all are at par with each other. Zn application showed beneficial effect on physiological process, plant metabolism, growth, there by leading to higher grain yield. The Zn application also enhanced the carbohydrates supply to kernels, increasing yield components like cob length, number of grains cob^{-1} and test weight, which was due to better vegetative growth of crop plant, which have direct influence on grain yield. Similar results were reported by Pokharel *et al.* (2009) [6], Kien *et al.* (2009) [5] and Jeet *et al.* (2012) [4].

Zn content and uptake

Zn content of maize grain is indication of potential yield response to applied Zn. Data presented in Table 3 showed that Zn content of grains. Zn concentration and uptake in grains increased in combined soil and foliar treatments of Zn application in comparison to Zn application through foliar. The highest Zn content and uptake was found in T_{14} treatment. Nutrient uptake (Zn) was vital in enhancing yield and nutrient content. Considerable increase in either nutrient content or in yield may increase the uptake. The uptake of Zn nutrient is a function of its improved metabolic reactions, activation of enzymes that leads to improvement in quality parameters as well as better harvest index. Similar results were reported by Pokharel *et al*. (2009) [6], Kien *et al*. (2009) [5] and Jeet *et al*. (2012) [4].

Table 3: Effect of methods and time of Zn application on Zn content and Zn uptake in maize (*Zea mays* L.)

Treatments		Zn content in grains (ppm)	Zn uptake (g ha^{-1})
Control (only recommended dose of NPK fertilizer)	T_1	3.88	18.41
Zn soil application (25kg/ha)	T_2	5.01	25.63
Zn Foliar at knee stage (1%)	T_3	4.32	20.99
Zn Foliar at tasseling (1%)	T_4	4.32	20.98
Zn Foliar at Grain filling (1%)	T_5	4.51	20.92
Zn Foliar at knee + tasseling (1%)	T_6	3.94	19.50
Zn Foliar at tasseling + Grain filling (1%)	T_7	4.24	21.90
Zn Foliar at knee + tasseling +. Grain filling (1%)	T_8	4.77	23.78
Zn_{soil} + Zn_{foliar} at knee stage (0.5%)	T_9	5.20	27.1
Zn_{soil} + Zn_{foliar} at tasseling (0.5%)	T_{10}	5.30	27.41
Zn_{soil} + Zn_{foliar} at Grain filling (0.5%)	T_{11}	5.40	27.88
Zn_{soil} + Zn_{foliar} at knee + tasseling (0.5%)	T_{12}	5.32	28.07
Zn_{soil} + Zn_{foliar} at tasseling +Grain filling (0.5%)	T_{13}	5.97	31.35
Zn_{soil} + Zn_{foliar} at knee + tasseling + Grain filling (0.5%)	T_{14}	6.26	32.96
CD (p = 0.05)		0.36	3.52

Conclusion

On the basis of present investigation, it may be concluded that soil+foliar application of Zn significantly influenced growth, yield and could be very helpful in improving grain Zn concentration and thus contributing to human nutrition (fortification). Foliar application of Zn at lateral stages i.e. grain filling increased Zn content in grains.

Future Line of Work

Due to extensive depletion of plant-available Zn in soils by cultivating high- yielding maize hybrids, application of Zn fertilizers or using Zn-fortified NPK fertilizers is an important practice for maize growers to maintain high yields and profitability.

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