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Effect of chelated iron and zinc application on growth and productivity of maize (*Zea mays* L.) in subtropical climate

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Abstract
The Iron (Fe) and Zinc (Zn) chelates were synthesized in laboratory using amino acid *viz*., glycine and methionine by excess alkali hydrolysis method used as micronutrient supplement for maize (*Zea mays* L.). The chelates were formulated in the ratio of 25:17 and 17:17. This study revealed the use efficiency of ferrous bisglycinate and zinc methionate compare over Fe and Zn fertilizer sources. The foliar application of formulation 17:17 at 0.2 per cent significantly increased the plant height and stem girth by 17% and 32% respectively over control. Similarly; the chlorophyll content and leaf area index also recorded 20% and 31% respectively over control. Foliar application of both formulations 17:17 and 25:17 at 0.2 per cent increased the overall growth and productivity of the crop by 21% over the control (T_1). When compared with sulphate salts (T_4 & T_8) and HEDP (T_5 & T_9) chelated iron and zinc source fertilizers the growth and productivity increased by 11% and 8% respectively. It concluded that formulations as foliar application has profound increased in the plant growth and productivity of the crop over the other Fe and Zn sources.

Keywords: Chelate, amino acids, fertilizer, micronutrient

Introduction

The micronutrients are playing a crucial role in growth and physiological process of plants and animals. Among the micronutrients, iron (Fe) and zinc (Zn) are the most prominent micronutrient deficiency occur in the world wide (Alloway 2009) [1]. The unavailability Fe and Zn micronutrient occur in soil condition such as high pH, high free calcium carbonate ($CaCO_3$), bicarbonate (HCO_3^-) ions, low organic matter content, high level of P content, antagonistic effects of other nutrients and adsorption of ions onto mineral surfaces (Singh 2008) [20]. The micronutrient availability improved by one of the smart way by chelation with chelating agents. The chelating agents or chelators are organic compounds that coordinate to the metal cations through the coordinate bonds in a ring structure. The structure of chelated complexes have higher stability, which avoids their precipitation, fixation, runoff in the soil thus increase their water solubility and plant availability (Sekhon 2003) [18]. Right now, chelated fertilizers are more efficient than the non-chelated fertilizer salts (Shivay *et al.*, 2016). In the recent decades, the chelating agents such as EDTA and DTPA were used to resolve the micronutrient deficient in the soil by their strong complexing capacity of element can increase the nutrient availability. Contrarily, it affects the soil by making complexation to the toxic heavy metals. Owing to their non-degradability of the synthetic chelating agent the toxicity persist in the soil over a period (Nortemann 1999 and Sillanpää 1997) [14, 19]. For while using amino chelated micronutrient fertilizer is the right source to make the nutrient availability for crop growth and also eco-friendly one (Dhanalakshmi *et al.*, 2019: Rukmani *et al.*, 2018) [3, 17]. In short, the study was aimed to evaluate the efficiency of soil and foliar applied Zn and Fe fertilizer sources *viz.*, HEDP and Sulphate form of fertilizer against the laboratory synthesized amino chelated fertilizers such as ferrous bisglycinate and zinc methionate on maize hybrid (CO MH 8) in alkaline soil.

Materials and Methods

A pot culture experiment was conducted during 2020 Rabi crop season (January to April) at green house, Department of Soils and Environment, Agricultural College and Research Institute, Madurai (Lattitude 9°58’ N and longitude 78°10’E). The climate is subtropical with average maximum temperature (T_{max} -32.5 °C) and minimum temperature (T_{min} -19.65 °C);

average relative humidity (RH-84%) and plants received optimum temperature for germination and growth.

Seeds of CO MH 8 (Maize hybrid) were sown in pots each contained 7 kg of alkaline (Fe and Zn deficient) soil collected from agricultural college. The recommended dose of fertilizer (250:75:75 Kg NPK ha^{-1}) in which quarter dose of N, full dose of P_2O_5 and K_2O were applied basally before sowing. Remaining half dose N at 25th DAS and quarter dose N at 45th DAS were applied in accordance with crop production guide issued by State Department (2020). The iron and zinc fertilizer sources were applied treatment wise at 30th and 45th day after sowing which were two critical stages of maize such as vegetative and knee high stages. Other agronomic practices were carried out homogenously to all the pots. The treatment pots were arranged in Completely Randomized Design (CRD) with five replications.

The iron and zinc chelated micronutrient mixture were synthesized in laboratory by excess alkali hydrolysis method whereas glycine and methionine were used as amino acid as chelating agents. In that synthesized amino acid chelated micronutrient mixtures were prepared in two formulations which had different iron: zinc ratios *Viz*., 25:17 and 17:17. The content of the formulations were analysed using Atomic Absorption Spectrophotometer and its physiochemical properties were analysed. The synthesized formulations were evaluated with HEDP and sulphate salt forms of iron and Zinc by the following treatments. The treatments imposed were T_1 -Absolute control, T_2-RDF + Foliar application of Fe and Zn formulation 25:17 at 0.2 per cent, T_3- RDF + Foliar application of Fe and Zn formulation 17:17 at 0.2 per cent, T_4- RDF + Foliar application of $FeSO_4$ at 1 percent + $ZnSO_4$ at 0.5 per cent, T_5-RDF + Foliar application of Fe-HEDP + Zn-HEDP at 0.2 per cent, T_6-RDF + Soil application of Fe and Zn formulation 25:17 at 7.5 kg ha^{-1}, T_7-RDF + Soil application of Fe and Zn formulation 17:17 at 7.5 kg ha^{-1}, T_8-RDF + Soil application of $FeSO_4$ at 50 kg ha^{-1} + $ZnSO_4$ at 37.5 kg ha^{-1}, T_9-RDF + Soil application of Fe-HEDP + Zn-HEDP at 5 kg ha^{-1}. The growth attributes were recorded at the three different stages viz., vegetative stage (30 DAS), tasseling stage (60 DAS) and at harvest stage (85 DAS). The plant height and stem girth were measured. The chlorophyll content was taken by using SPAD meter reading and Leaf Area Index (LAI) by the formula (McKee 1964) [11] using average leaf length and breadth of leaves. The yield attributes were recorded at harvest stage of the crop. The cob length and girth, number of seeds cob^{-1}, test weight were measured. The grain yield and stover yield were recorded on hectare basis. The harvest index (%) was also calculated. The data were subjected for statistical analysis using SPSS software (free version).

Results

The stable formulation mixtures were prepared and the physio-chemical properties were recorded as in the table 1. The nutritive content were analysed using atomic absorption spectroscopy.

Table 1: Physio-chemical properties of ferrous bisglycinate and zinc methionate formulations

Physio-chemical properties		**Ferrous bisglycinate: Zinc methionate (25:17)**	**Ferrous bisglycinate: Zinc methionate (17:17)**
A. Physical properties:			
Physical condition		Solid	Solid
Colour		Reddish brown	Light brown
Texture		Fine powder	Slightly coarser
Moisture percentage		4-5%	5-8%
Hygroscopicity		Slightly	Slightly
Water solubility		Highly soluble	Highly soluble
Organic solvent solubility		Insoluble	Insoluble
B. chemical properties:			
pH		7.3	7.5
Content (%)	Iron (Fe)	25	17
	Zinc (Zn)	17	17
	Nitrogen (N)	10.7	8.5
	Sulphur (S)	8.85	8.85

Growth Traits

The effect of different Zinc and Iron fertilizers on plant height at different stages (Table 2) were recorded. The measured plant height ranged between 78.40-95.70, 148.20-178.30 and 150.20-182.20 cm at vegetative, tasseling and harvest stages respectively. The plant height increased significantly (P = 0.05%) by foliar applied Fe and Zn formulation 17:17 at two per cent (T_3) and recorded the maximum height of 95.70, 178.30 and 182.80 cm at vegetative, tasseling and harvest stages respectively which was on par with foliar applied Fe and Zn formulation 25:17 at 0.2 per cent (T_2). The soil applied Fe and Zn formulation 17:17 at 7.5 kg ha^{-1} (T_7) also significantly (P = 0.05%) increased the plant height of 92.20, 169.6 and 173.6 at three different stages respectively in relation to foliar and soil applied HEDP and sulphate salts and the absolute control (T_1) recorded the lowest plant height of 78.2, 150.9 and 153.9 cm at three different stages respectively.

Table 2: Effect of different Zinc and Iron fertilizer sources on growth attributes viz., plant height and stem girth at different stages of hybrid maize

(Mean of five replications)

Treatment	**Plant height (cm)**			**Stem girth (cm)**		
	Vegetative stage	**Tasseling stage**	**Harvest stage**	**Vegetative stage**	**Tasseling stage**	**Harvest stage**
T_1	78.40	148.20	150.20	3.96	4.32	5.56
T_2	93.42	175.30	179.80	5.42	6.05	7.58
T_3	95.70	178.30	182.80	5.52	6.32	7.59
T_4	89.07	162.60	165.40	4.64	5.41	6.91
T_5	90.09	163.00	167.40	4.72	5.63	7.19

T_6	90.22	168.70	172.70	4.88	5.79	7.22
T_7	92.20	169.60	173.60	5.20	5.94	7.43
T_8	80.57	150.90	153.90	4.34	4.89	6.22
T_9	84.91	156.60	158.60	4.73	5.30	6.82
Mean	88.29	163.69	167.16	4.82	5.52	6.95
SEd±	1.692	3.078	4.058	0.115	0.107	0.162
CD (p=0.05)	3.53	6.46	8.52	0.242	0.226	0.341

The effect of different Zinc and Iron fertilizer sources on stem girth at different stages are presented in the Table 2. Significant differences were noticed with regard to stem girth. Stem girth ranged between 3.96-5.52, 4.32-6.32 and 5.56-7.59 cm at vegetative, tasseling and harvest stages respectively. It clearly showed that the girth was increased over different stages to different treatments. Foliar applied Fe and Zn formulation 17:17 at 0.2 per cent (T_3) resulted in highest stem girth of 5.42, 6.05 and 7.58 cm at cm at vegetative, tasseling and harvest stage respectively which was on par with the foliar applied Fe and Zn formulation 25:17 at 0.2 per cent (T_2). Furthermore, soil applied Fe and Zn formulation 17:17 at 7.5 kg ha^{-1} (T_7) had recorded higher stem girth of 5.20, 5.94 and 7.43 cm at different stages than that of absolute control (T_1) with the lowest of 3.96, 4.32 and 5.56 at three different stages.

Physiological Traits

The effect of different zinc and iron fertilizer sources on physiological traits (Table 3.) such as chlorophyll content and leaf area index at different stages of crop were increased in all the treatments except in the absolute control pots. However, both foliar and soil applied Fe and Zn formulations 25:17 and 17:17 greatly influenced the physiological traits. The chlorophyll content at three different stages recorded the maximum of 47.33, 50.50 and 55.70 respectively by foliar applied Fe and Zn formulation 17:17 at 0.2 per cent (T_3) which was on par with the foliar applied Fe and Zn formulation 25:17 at 0.2 per cent (T_2). The soil applied Fe and Zn formulation 17:17 at 7.5 kg ha^{-1} (T_7) recorded about 46.71, 48.8 and 54.79 respectively at different stages of crop. The leaf area index was also significantly affected by foliar applied Fe and Zn formulation 17:17 at 0.2 per cent (T_3) recorded the highest of 1.11, 4.03 and 5.67 at vegetative, tasselling and harvest stage respectively compared to HEDP chelate (T_5 & T_9) and sulphate salts (T_4 & T_8). Absolute control recorded the lowest of 0.63, 3.12 and 4.02 at three different stages respectively. The data clearly shown that the foliar and soil applied Fe and Zn formulation 25:17 and 17:17 had profound increased in physiological traits than other HEDP chelate and sulphate forms of fertilizer.

Yield Traits

The effect of different Zinc and Iron fertilizer sources on yield traits viz., cob length, cob girth, No. of seeds cob^{-1}, test weight and dry matter production (Table 4) were recorded. From the perusal of the data, it was evident that significant (P=0.05) increase in the cob length (24.8 cm), cob girth (19.20 cm), No. of seeds cob^{-1} (391), test weight (36.74 g) and dry matter production (14013 kg ha^{-1}) with foliar applied Fe and Zn formulation 17:17 at 0.2 per cent (T_3) which was on par with the foliar applied Fe and Zn formulation 25:17 at 0.2 per cent (T_2).

Table 3: Effect of different Zinc and Iron fertilizer sources on chlorophyll content and Leaf Area Index at different stages of hybrid maize

(Mean of five replications)

Treatment	Chlorophyll content			Leaf Area Index (LAI)		
	Vegetative stage	Tasseling stage	Harvest stage	Vegetative stage	Tasseling stage	Harvest stage
T_1	39.81	40.19	41.30	0.63	3.12	4.02
T_2	47.20	50.10	55.20	1.08	3.94	5.44
T_3	47.33	50.50	55.70	1.11	4.03	5.67
T_4	43.47	45.70	48.10	0.83	3.54	4.89
T_5	44.27	47.80	50.14	0.87	3.58	4.93
T_6	45.93	48.60	52.42	0.91	3.60	4.95
T_7	46.71	48.80	54.79	0.95	3.64	4.99
T_8	41.99	42.34	44.46	0.71	3.42	4.65
T_9	42.49	44.65	46.88	0.79	3.48	4.83
Mean	44.36	46.52	49.89	0.88	3.59	4.93
SEd±	1.067	0.942	0.94	0.021	0.067	0.094
CD (p=0.05)	2.24	1.97	1.97	0.04	0.14	0.19

However foliar application of Zn-HEDP + Fe-HEDP each at 0.2 percent (T_5) and foliar application of Zinc Sulphate at 0.5 percent + Iron Sulphate at 1 percent (T_4) were on par with each other. Regarding with soil application Zn-HEDP + Fe-HEDP each at 5 kg ha^{-1} (T_9) and soil application of Zinc Sulphate at 37.5 kg ha^{-1} + Iron Sulphate at 50 kg ha^{-1} (T_8) had recorded the lower yield than both formulations 25:17 and 17:17 but not than absolute control (T_1) and it recorded the lowest cob length (18.1 cm), cob girth (14.4 cm), No. of seeds cob^{-1} (286), test weight (33.11 g) and dry matter production (9938 kg ha^{-1}).

Table 4: Effect of different Zinc and Iron fertilizer sources on yield traits viz., cob length, cob girth, No. of seeds cob^{-1}, test weight and dry matter production of hybrid maize (CO MH 8)

(Mean of five replications)

Treatment	Cob length (cm)	Cob girth (cm)	No. of seeds cob^{-1}	Test weight (g)	Dry matter production (kg ha^{-1})
T_1	18.10	14.40	286	33.11	9938.00
T_2	23.50	18.90	384	36.26	13564.00
T_3	24.80	19.20	391	36.74	14013.00
T_4	21.00	16.60	340	34.83	11913.00
T_5	21.40	16.90	348	35.13	12130.00
T_6	22.30	17.70	364	35.66	12778.00
T_7	22.70	18.10	372	35.98	13006.00
T_8	19.20	15.10	303	33.64	10812.00
T_9	20.10	15.90	322	34.27	11263.00
Mean	21.46	16.98	345	35.07	12157.44
SEd±	0.417	0.439	7.252	0.67	290.90
CD(p=0.05)	0.87	0.92	15.23	1.4	630.08

Grain and Stover Yield

The effect of different Zinc and Iron fertilizer sources on grain and stover yield (Table 5) were recorded. The results revealed that grain yield of maize differed significantly with different sources of Iron and Zinc fertilizers both in soil and foliar application along with RDF. Treatment which received foliar applied Fe and Zn formulation 17:17 at 0.2 per cent (T_3) significantly (P=0.05%) recorded the highest grain yield of 5932 kg ha^{-1}. However, it was statistically on par with the treatments received with foliar applied Fe and Zn formulation 25:17 at 7.5 kg ha^{-1} (T_2) also recorded higher grain yield of 5763 kg ha^{-1}. Similarly, the stover yield of maize was also significantly (P = 0.05%) increased in the treatment received foliar applied Fe and Zn formulation 17:17 at 0.2 per cent (T_3). Furthermore, soil applied chelated micronutrient formulation 17:17 at 7.5 kg ha^{-1} (T_7) also had higher stover yield of 9097 kg ha^{-1} rather than other treatments. From the result it observed that foliar and soil applied Fe and Zn formulation 17:17 and 25:17 had better performance in increasing the grain and stover yield than the sulphate salts and HEDP chelated sources of Iron and Zinc fertilizers.

Table 5: Effect of different Zinc and Iron fertilizer sources on grain yield, stover yield and harvest index of hybrid maize

(Mean of five replications)

Treatment	Grain yield (kg ha^{-1})	Stover yield (kg ha^{-1})	Harvest index (%)
T_1	3911.00	7540.00	51.86
T_2	5763.00	9483.00	60.78
T_3	5932.00	9572.00	61.99
T_4	4771.67	8546.00	55.85
T_5	5004.67	8689.00	57.69
T_6	5287.33	9042.00	58.47
T_7	5455.33	9142.00	59.67
T_8	4198.67	7920.00	53.03
T_9	4495.67	8211.00	54.76
Mean	4979.3	8682.78	57
SEd	177.65	187.25	1.50
CD(p=0.05)	247.18	393.40	3.16

Discussion

Growth traits, the plant height and stem girth are most important traits which determine the vigour and potential of the maize crop. The Stem girth can directly influenced the carbon storage and its subsequent utilization for grain filling in maize. These are cases where the utilization of stem reserves for grain filling in irrespective of environmental conditions. The plant height and stem girth was significantly affected over the control at all stages of crop growth. The plant height and stem girth were increased due to the application of amino acids as chelating agents and made the nutrient available to the plants. The zinc activate of auxin metabolism leading to accelerate meristematic tissues at the shoot apex which in turn increased the height of the plants. However, excessive inactivation of auxin in the absolute control treatment leads retardation of stem elongation. Increased plant height might be also attributed to the intermodal distance as reported by Haghi *et al.*, (2006) [6]. Similar findings and observations reported by Eteng (2017) [5]. Also the stem girth can increased due to the carbon source from the amino acid chelated fertilizers whereas the stem acts as a reservoir of it. The report of the plant height and stem girth was same as reported by Pagad *et al.*, (2018) [15].

The chlorophyll content and Leaf Area Index (LAI) are in direct relationship with absorption of light energy from the sun. At different growth periods, application of chelated micronutrient mixture make the nutrient availability by chelation of nutrient besides supply nitrogen source from it which favours the crop to put forth more leaf area and also additional nitrogen and improve the activity of auxin, production of carbohydrates and organic compounds leading to accelerate meristematic activity at the shoot apex which in turn increased the leaf area. The increase in the chlorophyll content was due to the increased uptake of Iron from the amino acid chelated fertilizer whereas the iron can also directly involved in the synthesis of chlorophyll content in the leaves. Similar finding was reported by Durgude *et al.*, (2014) [4]. Tryptophan amino acid and IAA (Indole Acetic Acid) hormone are the two main factors for leaf area expansion which ultimately increased the leaf are index (Nadergoli *et al.*, 2011) [12].

The significant increase in yield attributes viz., cob length, cob girth, number of seed per cob and test weight may be due to the supply of amino acid chelated Iron and Zinc fertilizers as foliar spray rapidly taken by the plants. The Iron and Zinc directly involved as part of photosynthesis, assimilation and translocation of photosynthates from source part (leaves) to sink part (cob). The similar finding was observed by the Naveenaa *et al.*, (2018) [13].

Mahapatra *et al.*, (2018) [10] also reported that the micronutrient application could influence the nutrient uptake, utilization and also translocation of food materials to reproductive part of plant.

The increased dry matter production usually associated with higher leaf area per plant might be due to the improved nutrient availability through chelated micronutrient mixture and to increase in crop growth, photosynthetic activity and carbohydrate supplement to crop resulted in higher dry matter production. The results were evidenced with earlier findings of Parasuraman (2008) [16].

The higher grain and stover yield of maize hybrid was due to the higher growth attributing traits like plant height, stem girth, leaf area index and the physiological trait like chlorophyll content Khalid *et al.*, (2013) [8]. Kannan *et al.*, (2014) [7] also reported that multi-micronutrient mixture enhance the plant growth parameters and seed yield. The higher dry matter production and translocation of photosynthates from source to sink can contribute to higher yield. In addition Fe and Zn applied as foliar can directly took by the plant due to lower molecular size of amino acid chelate which involved in the various metabolic activities and enhanced carbohydrate synthetase. Similar findings were reported by Kumar *et al.*, (2018) [9].

The results obtained from the soil and foliar application of Iron and Zinc sources in the pot culture experiment, the foliar applied amino acid chelated micronutrient fertilizers in maize hybrid marked increase in the plant growth attributes, yield and yield attributes when compared to the other treatments because the amino acid chelated fertilizer shown the higher penetration rate than HEDP and sulphate form of fertilizers due larger molecular size of these synthetic chelates than amino acid chelated micronutrients and also amino acids from the chelates which can directly took as plant source, whereas derived as a nitrogenous source for the crop plants. Application of micronutrients in associated with NPK showed the better in growth and yield attributes. Therefore foliar applied Fe and Formulations 17:17 and 25:17 at 0.2 per cent with Recommended Dose of Fertilizer (250:75:75 kg ha^{-1} of N: P_2O_5: K_2O) surpassed all other Fe and Zn sources of fertilizer treatments by giving higher values for plant height, stem girth, chlorophyll content, Leaf Area Index (LAI), cob length, cob girth, number of seeds per cob, test weight, grain yield and stover yield.

Finally, our findings accept the hypothesis indicating that both foliar and soil applied Fe and Zn formulations 17:17 and 25:17 are more efficient fertilizer source than sulphate salts and HEDP chelated forms of fertilizers. Apart from that, usage of amino acid chelated fertilizers has no ill effects on soil physical, chemical, biological properties rather than synthetic chelating agents.

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