International Journal of ChemTech Research
CODEN (USA): IJCRGG ISSN: 0974-4290
Vol.8, No.10 pp 45-52, 2015

Chemical and physiological response of maize to salinity using cobalt supplement

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Abstract: Studies were carried out to investigate the efficiency of cobalt concentration (0,15, 17.5, 20, 22.5 and 25 ppm) in two maize cultivars (Giza 310 and Hi tick 2030) maize crop at private farms in El-hamoul Kafer El-Sheikh Governorate, Delta, Egypt, during 2013 and 2014 seasons. Increasing cobalt levels from 15 to 17.5 and 20 ppm significantly increased growth parameters of maize (plant height, leaves area, root length, shoot length and root fresh and dry weights) and shoots endogenous hormones such as Auxins (IAA), Gibberelines (GA_S) and Cytokinines (CY). While, abscisic acid (ABA) was gradually increased as cobalt levels in plant growing media was increased. Results also indicated that decreasing in SPAD value and Fe content in new leaves with increasing cobalt levels from 0.0 to 25.0 ppm. Application of 20.0 ppm cobalt produced the highest values of ear length, ear diameter, ear weight, ears grain weight /plant, 100-kernel weigh and grain yield (ton/fed). Data also revealed that, under saline condition, all used levels of cobalt significantly increased the content of macronutrients (N, P and K) as well as micronutrients (Ca, Mg, Mn, Zn and Cu) of maize grains compared with untreated plants. The highest significant figures of N, P, K, Ca, Mg, Mn, Zn and Cu were obtained by using cobalt at 20.0 ppm. Increasing cobalt in plant media more than 20.0 ppm decreased this promotive effect. Moreover, increasing cobalt concentration in saline plant media resulted in a progressive depression effect on Na, Cl and Fe content in the grains of maize. It could be concluded that using cobalt at the rate of 20 ppm resulted in increment of growth, yield and its quality of maize.
Keywords: Maize, Cobalt, Growth, Yield and attributes.

Introduction

Abiotic stresses have been recognized as the most important targets of crop improvement programs. There are relatively few ‘‘stress free’’ areas where crops may approach their potential yields. Abiotic environmental factors (abiotic stresses) are considered to be the main source (71%) of yield reductions. The estimation of potential yield losses by individual abiotic stresses are estimated at 17% by drought, 20% by salinity, 40% by high temperature, 15% by low temperature, and 8% by other factors.

Soil salinity is a major problem for agriculture throughout the world. The major constraints for plant growth and productivity are ion toxicity with excessive uptake of mainly Cl^- and Na^+ as well as nutrients imbalance caused by disturbed uptake of essential mineral nutrients[1]. Living with salinity is the only way of sustaining agricultural production in the salt affected soil[2]. So that, it is must to find the best management to alleviate salt hazard. [3] reported that in Egypt the area of irrigated land that is salt-affected is 33%. Salinity is a complex environmental constraint that presents two main components due to the decrease in the external osmotic potential of the soil solution and an ionic component linked to the accumulation of ions which becomes toxic at high concentrations.

Salinity is one of the major environmental factors limiting plant growth and productivity. Excess salt in the soil may adversely affect plant growth either through osmotic inhibition of water uptake by roots or specific ion effects. Specific ion effects may cause direct toxicity or, alternatively, the insolubility or competitive absorption of ions may affect plant nutritional balances[4]. Salt stress has toxic effects on plants and lead to metabolic changes, like loss of chloroplast activity, decreased photosynthetic rate and increased photorespiration rate which leads to an increased reactive oxygen species (ROS) production[5]. Salinity continues to be one of the world's most serious environmental problems as elevated levels of NaCl are naturally present in many agricultural fields. Global scarcity of water resources and the increase salinization of soil and water are reducing agricultural productivity. Crop performance may be adversely affected by salinity as a result of nutritional disorders. These disorders may derive from the effect on nutrient availability, competitive uptake, transport or partitioning within the plant[4]. Therefore, the development of methods and strategies to ameliorate the deleterious effects of salt stress on plants has received considerable attention. Enhancing stress tolerance in plants has major implications in agriculture and horticulture[6].

Maize is one of the most important strategic cereal crops in Egypt and the world. Increasing grain yield per unit area and increasing the maize cultivated area are recognized as a better solution to solve the gap between consumption and production. Therefore it's important to increase the maize yield. To overcome such deficiency, production per unit area must be maximized through good achievement of some agricultural practices.

Cobalt is considered to be beneficial element for higher plants in spite of the absence of evidence for direct role in their metabolism. This is true in spite of essentiality for photosynthetic activities of lower plants such as *euglena gracilis*. Cobalt is an essential element for certain micro-organisms particularly those fixing atmospheric nitrogen, its deficiency seem to depress the efficiency of nitrogen fixation[7]. Cobalt is an essential element for the synthesis of vitamin B_{12} which is required for human and animal nutrition[8]. Cobalt does not accumulate in human body, as other heavy metals with the increase in age. The daily cobalt requirements for human nutrition could reach 8 ppm depending on cobalt levels in the local supply of drinking water without health hazard[9]**.** [10] pointed that cobalt was used to reduce the harmful effect of salinity on tomato plants, transpiration rate being reduced. [11] showed that cobalt reduce the salinity and / or ethrel injury on tomato plants, a suggestion being introduced for possible use of cobalt with transplant irrigated with saline water to overcome the salinity hazard. Under salinity condition, [12] found that cobalt increased water content in pea plants. [13] stated that cobalt maximized the tolerance of wheat plants to soil salinity, increasing cobalt level in the cultural media up to 15 ppm cobalt stimulated growth, yield and quality as well as nutritional status (N, P, K, Ca, Mg, Zn, and Cu) in wheat shoots while Na and Cl concentration were decreased. [14]reported that growth parameters, pigment content, biochemicals and mineral content increase at 50 mg Co kg^{-1} soil when compared with the control. Further increase in the Co levels (100-200 mg kg-1 soil) has a negative effect on all the above parameters. Also, cobalt application increased wheat yields compared with control, and the greatest enhances were Co 1.05kg/hm2. Lower concentration of cobalt (0.45—0.75kg/hm2) improved grain quality (crude protein content, sedimentation value, wet gluten content, dry gluten content and valorimeter value) and higher addition of Co (over 1.35kg/hm2) resulted decrease the promotive effect in values of grain quality[15, 16] .

The present investigation was executed with an objective to study the effects of Cobalt supplement on growth, yield, nutrients content and biochemical constituents of *Zea mays* L under saline condition.

Material and Methods

Two field experiments were conducted during the 2013 and 2014 growing seasons at the Experimental a private Farm at El-hamoul, Kafer El-Sheikh Governorate, Delta, Egypt, to study the response of maize yield and its components to different concentration of cobalt under saline condition. The experiments were established with split- plot design using four replicates. The main plots included the two maize cultivar (Giza 310 and Hi tick 2030), while the sub-plots occupied by cobalt concentration (0, 15.0, 17.5, 20.0, 22.5 and 25.0). The experimental unit area was 10.5 m^2, contained 5 ridges (3.0 m length and 0.7 m apart). The chemical and physical analysis of the used soil during the two growing seasons are illustrated in Table (1).

Grains of maize were drilled in one side of ridge in hills 25 cm apart at a rate of 10 kg/fed. The sowing date was May 8th and 12th in the 1st and 2nd seasons, respectively. At 25 days after sowing, plants were thinned to secure one plant/hill followed by the first irrigation. Phosphorus fertilizer in the form of calcium super phosphate, 15.5 % P_2O_5 was applied during the soil preparation at the rate of 100 kg/fed. Ammonium sulfate (21% N) was applied at 120 kg/fed at four times during the growing season. All other recommended cultural practices were adopted throughout the two growing seasons. The seedlings (at the third truly leaves) were irrigated once with cobalt concentrations (control, 15.0, 17.5, 20.0, 22.5 and 25.0 ppm) as cobalt sulphate form.

Table (1): Some physical and chemical properties of the used soil.

Physical	Particle Size distribution (%)			Soil texture class		Field capacity			Welting point			Available water	
	Sand	Silt	Clay			%							
	19.25	15.64	65.11	Clay		46.2			15.9			30.3	
Chemical	PH^a	EC^b		Soluble cations (meq/L)					Soluble anions (meq/L)				
				Ca^{++}	Mg^{++}		K^+	Na^+	Hco_3^{--}	Co_3^-		Cl^-	So_4^{--}
	8.6	12.5 d/sm		47.5	23.7		0.15	167	0.80	-		178	69.5
Total available (mg/100g)			Available micronutrients (ppm)				CEC Meq/100 g soil	ESP	Cobalt (ppm) Soluble	Available	Total	$Caco_3$	OM
N	P	K	Fe	Mn	Zn	Cu	58.3	27.2	0.34	4.46	16.9	%	
3.2	3.0	18.1	75.0	7.0	10.0	5.0						2.71	1.20

Measurements:

Vegetative growth: After 80 days from sowing, all growth parameters of maize plants such as plant height, number of leaves, leaves area, root length as well as shoot and root fresh and dry weights were recorded according to[17].

Determination of Endogenous Hormones: Auxins, Gibberelines, cytokinins and abscisic acid in maize fresh shoots were determined according to[18].

Determination of Proline Content: Fresh leaves sample (5 g) were taken for this determination according to[19].

Determination of SPAD value: The total chlorophyll content (SPAD value) of flag leaf was determined by chlorophyll meter (SPAD-502 plus) according to soil plant analysis department section, Minolta Camera Co., Osaka, Japan as reported by[20].

Yield and its components: At harvest, ten guarded plants were chosen randomly from each experimental unit to measure ear length, ear diameter, number of rows /ear, ear weight and 100-kernel weight. Moreover, whole plants of each experimental unit were harvested to estimate grain yield (ton /fed).

Measurement of Nutritional Contents: Macronutrients (N, P and K) and micronutrients (Fe, Mn, Zn and Cu) as well as cobalt of maize grains were determined according to[21].

Measurement of Chemical Constituents: The concentrations of the studied chemical contents percentages (oil, protein, starch, total soluble sugars and total phenol) in maize grains were determined according to[22].

Statistical Analysis: All the obtained data from each season were exposed to the proper statistical analysis of variance according to[23].

Results and Discussion

Vegetative growth :

After 41 days from sowing the plants of Hi tick 2030 cultivar was burn as a result of salinity stress. Application of cobalt at 20 ppm/fed gave the greatest values of plant height, leaves area, root length along with shoot and root fresh and dry weights of Giza 310 cultivars (Table 2). Increasing cobalt level more than 20 ppm reduce the promotive effect. Vice- versa untreated plots by cobalt recoded the lowest values of the previous characters. These observations are consistent with previous reports obtained by[24]. They reported that the lower doses of cobalt resulted in maximum growth and yield of tomato plants as compared with the higher ones. They reported that responses associated with low cobalt levels may be attributed to catalase and peroxidase activities which were found to decrease with low levels of cobalt and increase with the higher ones. These enzymes are known to induce plant respiration, so superior resulting in successive consumption for products of photosynthesis and consequently reduced in plant growth. Moreover, low cobalt levels being with positive effect due to several induced effects in hormonal synthesis and metabolic activity, while the higher cobalt levels were found to increase the activity of some enzymes such as peroxidase and catalase in plant and hence increasing the catabolism rather than the anabolism. Cobalt help plants to resist stresses caused with high salinity[25].

Endogenous hormones:

Data presented in Table (3) showed that, all cobalt treatments had increased shoots endogenous hormones such as Auxins, Gibberelines, Cytokinines and Abscisic acid in maize shoots grown under salinity condition compared with control. Increasing cobalt levels up to 20.0 ppm significantly increased these hormons. Cobalt at 20.0 ppm recorded the highest maize phytohormones. However, higher cobalt levels above 20.0 ppm has reduce the positive effect in endogenous maize hormones due to the effect of stress salinity. Endogenous plant hormones such as Auxins, Gibberelines, cytokinines and Abscisic acid are known to be involved in the regulation of plant response to salinity stress and counteract the adverse effect of stress conditions. Plant hormones as natural products, can stimulate physiological response to high soil salinity. Different strategic crops are being employed to maximize plant growth under saline conditions. In fact, the levels of Auxins, Gibberelines, Cytokinines hence plant growth and help plants to resist stresses, while Abscisic acid play a central role in the hormonal control of in the hormonal control of water balance, reduction in the values of transpiration noticed due to cobalt addition could be attributed to content and decreased leaf water deficit and help plants to tolerate the salinity stress. These data are in harmony with those obtained by[26, 11] they showed that cobalt increase cytoplasmic osmotic pressure, leaf resistance to dehydration and decreased the willing coefficient of pea plants . and Abscisic acid under salt stress are help plants to tolerate the high salinity soil. These results are in harmony with those obtained by[27] who found that the effect of applied IAA and GA_S in alleviation of salt stress might be through the activation of a specific enzyme, which participates RNA and protein synthesis. [12] added that Abscisic acid was gradually increased as the rate of applied cobalt was raised in the growth media. Abscisic acid help plant to tolerate the salinity stress. Abscisic acid is an energy inhibitor which results in inhibition for potassium translocation to the gard cell and subsequently stomata closer. Also, Abscisic acid could modify the plants water economy before the leaves become stressed[28].

Leaf water potential:

Data presented in Table (3) indicated that, addition of different cobalt levels (0.0, 15.0, 17.5, 20.0, 22.5 and 25.0 ppm) in plant growth media (El-Hamoul saline soil) significantly decrease maize leaf water potential. These results are in harmony with those obtained by[29] who found that the observed beneficial effect of cobalt on growth of barley plants on salinized could be increase in the leaf water potential relative to compared those untreated with cobalt. The leaf water potential could enhance the photosynthesis process directly by influencing the photosynthesis system or indirectly by decreasing the total leaf resistance to the diffusion of Co_2 into the leaf.

Table (2) : Effect of cobalt on vegetative growth of maize cultivars.

Cobalt treatments (ppm)	Plant height (cm)	Leaves no. /plant	Leaves area (cm^2)	Length of root (cm)	Fresh weight (g)		Dry weight (g)	
					Root	Shoot	Root	Shoot
	Giza 310							
Control	256	13	3868	9.4	796	175	212	21.8
15.0	284	13	4304	10.2	1018	202	247	28.3
17.5	292	13	4310	11.9	1127	226	267	31.1
20.0	305	14	4977	14.0	1214	241	285	36.2
22.5	302	14	4965	12.6	1203	237	281	35.1
25.0	297	14	4308	12.0	1141	232	279	32.3
LSD 5%	3.0	NS	15.0	0.6	11.0	4.0	2.0	1.2
	Hi tick 2030							
Control	78	9						
15.0	110	9						
17.5	115	9						
20.0	121	9						
22.5	120	9						
25.0	119	9						
LSD 5%	2.1	NS						

Proline content:

Averages of Proline content was appreciably influenced by cobalt levels in combined both seasons as shown in Table (3). In this respect, with each increase in cobalt level there was a progressive increase in Proline content. Application of cobalt at 25.0 ppm recorded the highest value of Proline content. On the other side, the lowest Proline content was observed with untreated plants. The obtained results confirmed with those previously discussed by[24] on tomatoes. She found that the salt stress effect increased praline content. Cobalt help tomato plant to resist stress caused by high salinity. The vital role of cobalt in porline biosynthesis, in modifying the plant water economy in tomato leaves of both varieties was confirmed.

Chlorophyll and iron content in leaves:

Presented data in Table (3) clearly indicate that in maize lower old leaves, increasing cobalt levels in plant growing media insignificant effect on both chlorophyll and iron content. The content of chlorophyll and iron in lower leaves higher than maize upper ones. In upper new leaves, control plants record the lowest chlorophyll content as salinity stress. While, the content of iron under salinity condition was increase with the increasing of cobalt in plant growing media, as unexpected and as mentioned by[30]. They found that cobalt addition in plant media in progressive depression effect on iron status in soybean and explained on basis of certain antagonistic relationships between cobalt and iron. [31] pointed out that cobalt at 50 mg/ kg soil increased photosynthetic pigments as chlorophyll a, chlorophyll b and total chlorophyll content in groundnut leaves. [13, 32] added that cobalt at 50 mg/kg soil increased photosynthetic pigments as chlorophyll a, chlorophyll b and total chlorophyll content in both maize and green gram leaves.

Table (3) : Effect of cobalt on hormones content, leaf water potential, proline content, chlorophyll and iron content of corn plants.

Cobalt treatments (ppm)	Hormones content (µg/g^{-1} fresh weight)				Leaf water potential (-bar)	Proline content g/100g fresh leaves	SPAD value		Iron content	
	Auxins	Gibber-elines	Cytok-inines	Abscisic acid			In new leaves	In old leaves	In new leaves	In old leaves
Control	1.198	1.395	1.169	----	-19.5	0.28	39.9	49.5	24.1	27.5
15.0	1.276	1.940	1.414	0.978	-17.0	0.34	41.2	49.6	24.1	27.5
17.5	1.885	2.602	2.122	1.574	-14.9	0.37	43.5	49.6	23.0	27.2
20.0	2.212	2.936	2.489	1.972	-11.5	0.41	43.8	49.7	22.4	27.5
22.5	2.150	2.722	2.469	2.256	-13.7	0.46	44.0	49.8	21.0	27.1
25.0	2.112	2.319	2.426	2.703	-16.2	0.50	44.5	49.9	19.7	27.9
LSD 5%	0.004	0.103	0.058	0.153	0.80	0.70	2.3	NS	0.2	NS

Yield and its components:

Results in Table (4) revealed that cobalt concentration had markedly affected on ear length, ear diameter, number of rows /ear, ear weight, 100-kernel weigh, ears grain weight /plant and grain yield (ton/ha). It is obvious that addition of 20.0 ppm cobalt produced the highest values of aforementioned traits while using untreated plots gave the lowest values previous characters. Increasing cobalt concentration of 15.0, 17.5 and 20.0 ppm gave higher values of grain yield ton/ha. They significantly increased grain yield ton/ha over the untreated check by 19.56, 27.71 and 31.11% , respectively. On the other hand, cobalt concentration had insignificant effect on number of rows/ ear. Increasing cobalt rate more than 20 ppm significantly decrease the promotive effect. [13, 32] who found that cobalt addition in soil increased all growth parameters along with yield parameters such as seedling vigour, number and weight of pods and seeds yield / plant in green gram (*Vigna radiate* L.) and maize (*Zea maize* L.) plants. These data are in harmony with those obtained by[33] who stated that the lower doses of cobalt resulted in maximum growth and yield of tomato and cucumber plants as compared with the higher ones. They reported that responses associated with low cobalt levels may be attributed to catalase and peroxidase activities which were found to decrease with low levels of cobalt and increase with the higher ones. These enzymes are known to induce plant respiration, so superior resulting in successive consumption for products of photosynthesis and consequently reduced in plant growth. Moreover, low cobalt levels being with positive effect due to several induce the effects in hormonal synthesis and metabolic activity, while the higher cobalt levels were found to increase the activity of some enzymes such as peroxidase and catalase in plant and hence increasing the catabolism rather than the anabolism[24].

Table (4) : Effect of cobalt on yield and yield attributes as well as chemical contents of maize grains.

Cobalt treatments (ppm)	Yield and yield attributes						Chemical contents of grains %				
	Ear length (cm)	Ear diameter (cm)	number of rows /ear.	Weight of ears/ plant (gm)	100-kernel weigh (gm)	Grain yield (ton/ fed)	Oil	Protein	Starch	Total Soluble sugars	Total phenols
Control	15.1	4.13	13.12	241	27.73	1.559	3.11	6.74	41.1	10.3	0.78
15.0	17.2	4.69	13.42	271	29.32	1.864	3.68	7.02	43.9	10.8	0.97
17.5	18.5	4.95	13.65	294	30.91	1.991	3.79	7.63	44.4	11.2	1.68
20.0	19.3	5.32	13.92	319	31.87	2.044	4.06	8.37	45.5	12.0	1.79
22.5	18.7	5.20	13.85	314	30.53	2.021	4.06	8.15	43.6	11.9	1.74
25.0	18.5	5.01	13.57	312	30.41	2.015	4.01	8.03	42.5	11.4	1.62
LSD 5%	0.5	0.23	NS	5.0	0.51	0.15	0.05	0.13	0.8	0.5	0.15

Chemical constituents of grains:

Under salinity condition, considerable effects of cobalt concentration on some chemical contents in maize grains such as oil, protein, starch, total soluble solid and total phenols as shown in Table (4). Data clearly indicated that all cobalt levels significantly increase the studied chemical constituents compared with untreated plants. Increasing cobalt rate in plant growing media up to 20 ppm significantly increased the studied chemical contents. Cobalt at 20 ppm resulted the superior values. Data also reveled that increasing cobalt above 20 ppm significantly decrease the promotive effect. These results add more support to those reported by[34] who fond that cobalt levels significantly increased as oil, protein, starch, total soluble solid and total phenols in canola seeds compared with control. Cobalt at 12.5 ppm gave the highest figures. Increasing cobalt rate more than 12.5 ppm significantly decrease this positive effect.

Nutritional content of maize grains:

Presented data in Table (5) showed the effect of different cobalt levels on mineral composition of maize grains. Data revealed that, under saline condition, all used cobalt levels significantly increased the content of macronutrients (N, P and K) as well as micronutrients (Ca, Mg, Mn, Zn and Cu) of maize grains compared with untreated plants. The highest significant figures of N, P, K, Ca, Mg, Mn, Zn and Cu were obtained by using cobalt at 20.0 ppm. Data in Table (5) also, indicate that both Na and Cl resulted in a progressive depression as cobalt concentration in plant growing media increased. Increasing cobalt in plant media more than 20.0 ppm decreased this promotive effect. These data are in harmony with those obtained with[29] in barley under saline condition. Data in Table (5) also indicated that, increasing cobalt concentration in saline plant media resulted in a progressive depression effect on iron content in the maize grains. These data are agree with these obtained by[35] who showed certain antagonistic relationships between cobalt and iron. Data in Table (5) also, indicate that, increasing cobalt concentration in plant media increased cobalt content in maize grains as compared with untreated plants. Cobalt levels in maize grains of 7.88 ppm with the highest cobalt treatment (25.0 ppm) is below the dangerous level. [9] reported that the daily cobalt equipments for human nutrition could reach 8 ppm depending on cobalt levels in the local supply of drinking water without health hazard. Since the daily consumption of wheat grains does not exceed a few grains.

Table (5) : Effect of cobalt on elements contents of corn grains.

Cobalt treatments (ppm)	Macronutrients (%)							Micronutrients (ppm)				Co (ppm)
	N	P	K	Ca	Mg	Na	Cl	Mn	Zn	Cu	Fe	
Control	1.08	0.64	1.02	1.11	0.19	5.42	4.71	3.04	2.11	3.25	6.09	0.95
15.0	1.12	0.71	1.48	1.19	0.23	5.19	4.52	3.16	2.17	3.36	6.06	1.77
17.5	1.22	0.78	1.79	1.38	0.29	5.01	4.16	3.24	2.22	3.42	6.04	2.54
20.0	1.34	0.84	2.02	1.52	0.33	4.87	3.90	3.37	3.25	3.56	6.02	3.66
22.5	1.30	0.80	1.88	1.50	0.30	4.75	3.76	3.29	3.13	3.44	5.89	5.92
25.0	1.28	0.80	1.84	1.46	0.30	3.69	2.89	3.21	2.79	3.36	4.42	7.88
LSD 5%	0.11	0.04	0.4	0.02	0.03	0.12	0.65	0.2	0.19	0.23	0.12	0.21

Conclusion:

Cobalt at 20 ppm has a significant promotive effect on endogenous hormones, leaf water potential, Proline content, yield quantity and quality under salinity condition. Cobalt help maize plants to tolerate high soil salinity. Cobalt reduce the harmful effect of salinity on maize plants.

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