



MAXIMIZATION OF DROUGHT TOLERANCE OF BEAN PLANTS USING COBALT SUPPLEMENTATION: B- PHYSIOLOGICAL AND CHEMICAL CONTENTS IN PLANTS

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Abstract

A field experiment was carried out at Research and Production Station, National Research Centre, El- Noubaria region, Beheira Governorate, Delta, Egypt under three water regime (100%, 80% and 60%) condition, on 2017 season. Seedlings of green bean, Paulista variety at three truly leaf stage were irrigated once with 4, 8, 12, 16 and 20 ppm cobalt beside the control. All required agriculture managements for plants growth and production were carried out as recommended by the Ministry of Agriculture. The obtained results could be summarized in the following:-

- * All cobalt concentrations significantly increased endogenous Auxins and Gibberellins in bean plants compared with control.
- * As cobalt levels increased in plant media ,the values of water consumption decreased under different rates of irrigated water (100%, 80% and 60%).
- * The values of leaf water potential significantly increased with all cobalt rates compared with untreated plants .
- * The content of Abscisic acid gradual increase as water level decrease (80%) and (60%). Abscisic acid reduces plant water loss.
- * All cobalt concentrations resulted the promotive effect on total proteins, total carbohydrates , total soluble solids and vitamin "C" of bean pods under the studied water levels,
- * Cobalt at 12 PPM hence Bean Physiological and some chemical constituents under different levels of irrigated water. Cobalt help bean plants to tolerate water deficit up to 60% from its water requirements.
- * Cobalt increase water use efficiency and save 20% from Bean water requirements.

Keywords: Bean, Cobalt, Physiological contents, Chemical constituents.

Introduction

Green bean (*Phaseolus vulgaris* L.) is considered one of the important vegetable crops cultivated in Egypt. Bean plants are relatively sensitive to environmental conditions, mostly drought, (Mohamad AL Hassan et al., 2016).

Climatic changes could be summarized in the increase in temperature, global warming, the decrease of waters, increase in soil salinity floods and the change in the amount of rainfall, dryness, the increase in atmosphere humidity, the increase in ozone concentration, the pollution resulting from burning fuel and the excessive in the use of mineral fertilizers (Kang et al., 2009).

Water stress occurs due to water deficit, caused by high soil salinity or drought. In case of high salinity, water exists in the soil but plants cannot uptake it, which is called physiological drought (Chaves et al., 2003). When water is gradually lost from a completely saturated soil, first by draining freely under the effect of gravity, and the rate of loss progressively slows down till no more water drains away, the soil is called to be at field capacity. Water stress was associated with the bend neck phenomenon and mediated by ethylene whose production seemed to be correlated with status of endogenous Abscisic acid and presence of cobalt in the plant growth media.

Further decrease in water uptake by plant roots or by evaporation reduces the water content of the soil, till no more loss, a stage called the wilting point at which roots cannot absorb water important to meet their needs, and the plants wilt and die (Nagarajan and Nagarajan, 2010).

Cobalt, as one of the beneficial element, seems to have a positive effect in higher plants to withstand water stress

conditions. Egrove (2000) showed that soil application of cobalt (3 mg/kg dry soil) increased leaf water content and decreased water deficit during daytime in tomato and potato leaves. This application also, increased water absorption capacity and the content of strongly bound H₂O in the leaves. Cobalt increased cytoplasmic pressure and leaf resistance to dehydration and decreased the wilting coefficient of the plants, increasing there by their drought resistance. Soaking soybean seeds in cobalt solution partially alleviated the effect of moisture stress on seedling growth (Duan and Auge, 2001).

Anter and Nadia Gad (2001) studied the relationship between cobalt status, water consumption and hormonal balance under two levels of soil moisture. The anatomical features of plant root were also considered. The obtained results revealed that cobalt application reduced transpiration rate with all cobalt treatments. Low cobalt concentration has a promotive effect on leaf water potential and Abscisic acid, Auxine and Gibberellins as well as xylem and phloem were beneficially affected by cobalt, especially at water deficit condition.

One of the most striking effects of cobalt is its ability to improve water relations inside plants. Its addition has been shown to increase water absorption capacity and the content of strongly bound H₂O in plant leaves

Also Schautmann and Wenzel (2002) studied the effect of cobalt on *Phaseolus vulgaris* under water stress condition such as drought, water logging and salinity and found that cobalt increased both Abscisic acid and ethylene hormones which are known to enhance plant resistance, promote old leaves abscission and reduce plant water losses. Nadia Gad

and Atta-Aly (2006) reported that, high cobalt level (over 18 ppm) inhibited ethylene production on both tomato and cucumber plants. This was true in spite of the vital role of ethylene and Abscisic acid at low cobalt concentration in adjusting plant water balance and consequently plant growth.

Moreover, Nadia Gad (2006a) recommended the use of 7.5 PPM cobalt during the seedlings stage of tomato if grown in a newly reclaimed soil. This cobalt level has a promotive effect on plant growth, leaf water potential, Abscisic acid as well as xylem and phloem tissues being increased especially under water deficit condition.

Nadia *et al.*, (2018) Pointed that all cobalt treatments significantly increase the studied Bean growth and yield parameters along with minerals content under different irrigated water levels (100%, 80% and 60% compared with untreated plants. Cobalt at 12 PPM gave the greatest values. With cobalt at 12 PPM, green pods yield of plants which grown under 80% irrigated water (7.667 ton/fed) is no significant with those which grown under 100% one (7.886 ton/fed). Cobalt increase water use efficiency and save 20% from irrigated water.

Materials and Methods

Soil analysis:-

Soil samples were taken from El-Noubaria farm and such samples were air dried and then prepared for analysis using conventional techniques.

Physical analysis:-

Particle size distribution, saturation percentage curve, moisture characteristics curve, hydraulic conductivity, total porosity and texture class were determined according to (Blackmore 1972).

Chemical analysis:-

Electrical conductivity (ds/m-1), pH in soil- water suspension (1:2.5), organic matter content (%), CaCO₃ (%), cation exchange capacity, cations and anions in meq/liter (in soil paste), macro and micronutrients were determined according to (Black *et al.*, 1982).

Cobalt analysis:-

Total cobalt were determined in Aqua regia extraction (Cottanie *et al.*, 1982). The water soluble cobalt as well as available cobalt (DTPA extractable) was assayed according to Black *et al.*, (1982). Determination of cobalt was carried out using Atomic Absorption Spectrophotometer, Varian AA-20.

Experimental work:-

A field experiment were carried out at Research and Production Experimental Station of National Research Centre, Noubaria Site, El-Beheira Governorate to investigate the effect of three water rates (100, 80 and 60 %) from water requirement. Five Cobalt rates (4, 8, 12, 16 and 20 ppm cobalt beside control) within four replicates on bean plants. The area of experimental plot was 10.5m². Split plot design was used with three replicates, water regime treatments occupied the main plots, while application of cobalt were distributed randomly in the sub plots. Seeds of Bean, Paulista variety were sown at 15 March, 2017. Seedlings at the third truly leaf stage, were irrigated with cobalt sulphate once, with 4, 8, 12, 16 and 20 ppm cobalt beside the control. All required agriculture managements for plants growth and production were carried out as recommended by Ministry of Agriculture.

Measurement endogenous hormones:

After 55 days (Vegetative stage end), fresh samples of shoot were taken for analysis of endogenous Auxins, Gibbrillins, and Abscisic acid according to Shindy and Smith (1975).

Measurement leaf water potential:-

Using leaf water meter, leaf water potential were determined in plants fresh leaves during plant season.

Measurement water consumption rate:-

Transpiration rate under the different studied levels (100%, 80% and 60% from irrigated water was recorded with all cobalt concentrations.

Measurement of chemical constituents:-

After 90 days, in bean pods from sowing in bean pods, total proteins, total carbohydrates, total soluble solids, and vitamin "C" were determined according to A.O.A.C (1995).

Table 1: Physical and chemical analysis of the experimental soils.

Physical	Particle size distribution %			Soil Texture class	Water saturation	Field capacity		Wetling point	Available water		
	Sand	Silt	Clay	Sandy loam			%				
	69.8	26.7	3.5		20.0	14.4		3.9			10.5
Chemical	pH		EC	Soluble cations (meq/l)				Soluble anions (meq/L)			
	(1:2.5)		(ds/m)	Ca ⁺⁺	Mg ⁺⁺	K ⁺	Na ⁺	HCO ⁻	CO ₃	Cl ⁻	SO ₄ ⁼
	8.0		1.0	9.0	1.4	5.4	3.26	-	1.18	6.60	2.4
	Total	Available	Available				Cobalt (ppm)			CaCO₃	OM
	(ppm)		Micronutriments (ppm)				Soluble	Available	Total		
N	P	K	Fe	Mn	Zn	Cn	9.39	1.78	9.68	%	0.19
25.2	15.3	10.2	23.0	10.5	3.62	5.22				3.17	

Statistical analysis:-

All data were subject to statistical analysis according to procedure outlined by SAS (1996) computer program and

means were compared by LSD method according to Snedecor and Cochran (1982).

Results and Discussion

A Green common bean (*Phaseolus vulgaris* L.) is considered one of important vegetable crops cultivated in Egypt. Bean plants are relatively sensitive to environmental conditions, mostly drought, (Mohamad AL Hassan et al., 2016).

Physiological contents in plants:-

Endogenous hormones:-

Data presented in Table (2) indicate that, all cobalt levels significantly increased bean shoots endogenous hormones such as Auxins, Gibberellins and Abscisic acid compared with control. Cobalt at 12 PPM resulted the superior values of bean shoots phytohormones. Increasing cobalt rates up to 12 PPM significantly increased these hormones.

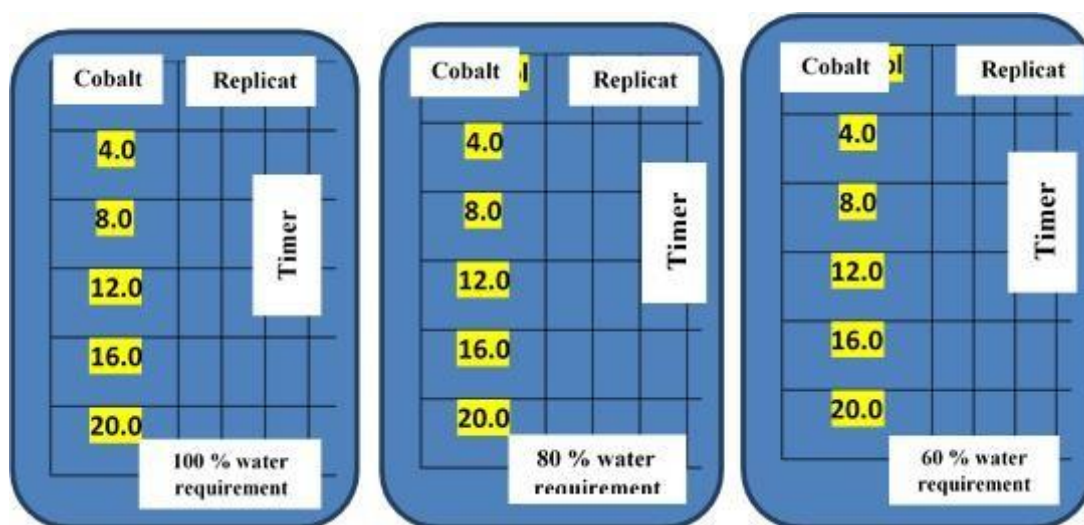


Fig. 1: Layout of field experiments at NRC Farm, Noubaria, Beheira Governorate

Table 2: Effect of cobalt on some endogenous hormones of bean shoots under different irrigated water levels.

Some endogenous hormones	Auxins ($\mu\text{g/g}$ fresh tissue)				Gibberellins ($\mu\text{g/g}$ fresh tissue)			
	Irrigated water (%)							
	Cobalt treatments (PPm)	100	80	60	Mean	100	80	60
Control	1.585	1.574	1.528	1.557	2.113	1.989	1.902	2.021
4	1.623	1.598	1.576	1.598	2.166	2.032	1.985	2.061
8	1.662	1.659	1.624	1.648	2.194	2.180	2.021	2.132
12	1.733	1.729	1.670	1.711	2.215	1.896	1.747	1.953
16	1.724	1.714	1.649	1.689	2.186	1.819	1.726	1.910
20	1.688	1.675	1.503	1.622	2.179	1.792	1.686	1.286
Mean	1.669	1.658	1.592		2.174	1.951	1.845	
LSD Cobalt (A)	0.8				0.13			
LSD water (B)	0.22				0.25			
LSD (AxB)	0.176				0.033			

However, higher cobalt concentration more than 12 PPM was decreased the promotive effect. Plant hormones are natural products, they stimulate physiological response in plant growth. Different strategic are being employed to maximum growth (Nadia Gad et al., 2018). These results are in harmony with those obtained by (Nadia Gad, 2005a) who stated that low cobalt levels (7.5 PPM) gave the positive effect due to several induced effects in hormonal synthesis and metabolic activity. While the higher rates increased the activity of some enzymes such as peroxidase and catalase in tomato plants and hence increasing the catabolism rather than anabolism. Confirm these results Basu et al., (2006) who found that the lower cobalt concentrations significantly increased the growth and yield of groundnut as compared with higher ones.

Water consumption:-

Table (3) show that the values of water consumption per plant were the highest in the cobalt untreated plants (control). Supplementing the plant media with cobalt,

generally reduced transpiration rate. As cobalt rate in plant media increased, the values of water consumption per Bean plant decreased under different irrigated water levels (100%, 80% and 60%). These results are in harmony with those obtained by Radiri (2004) who found that cobalt increased water content in cotton leaves.

Data in Table (3) clearly indicate that the values of transpiration was the highest in the Bean plants (control). Application the plant media with cobalt, generally, reduced transpiration rate. As cobalt concentration increased the values of transpiration decreased under different irrigated water levels (100%, 80% and 60%).

These results agree with those obtained by Angelove et al., (1993) who stated that cobalt increased water content in pea plants, the rates of transpiration process being decreased.

Leaf water potential (-bar):

Data in Table (3) reveal that as decreasing irrigated water treatment, leaf water potential in Bean leaves significantly increased. This may go along with

results of Luo *et al.*, (2016) they showed that water deficit mostly reduced cotton leaves water potential and in turn the leaf areas and leaf senescence could be observed under severe water stress.

Data in Table (3) show that the values of leaf water potential significantly increased with all cobalt levels compared with untreated plants. These finding are strongly supported by Egrove (2000) who showed that soil application of cobalt at 3 mg per kg dry soil increased leaf water content

and decreased water deficit during daytime in tomato and potato leaves. This application also, increased water absorption capacity and the content of strongly bound H₂O in the leaves. Cobalt increased cytoplasmic pressure and leaf resistance to dehydration and decreased the wilting coefficient of the plants, increasing there by their drought resistance.

Table 3: Effect of cobalt on some physiological contents of bean shoots under different irrigated water levels.

Some physiological content	Water consumption per plant (mm/season)				Leaf water potential (- bar)				Abscisic acid (µg per g fresh tissue)			
	Irrigated water (%)											
Cobalt treatments (PPm)	100	80	60	Mean	100	80	60	Mean	100	80	60	Mean
Control	510	408	306	408	-12.6	-12.2	-12.0	12.3	-	0.690	0.875	0.783
4	482	381	287	383	-11.2	-11.0	-10.7	11.0	-	0.844	1.506	1.18
8	460	364	256	360	-9.8	-9.5	-9.2	9.5	-	0.918	1.946	1.93
12	439	346	241	342	-8.3	-8.0	-7.8	8.03	-	1.225	2.490	1.86
16	422	339	234	332	-8.0	-7.3	-7.0	7.43	-	1.905	3.130	2.52
20	394	317	219	310	-7.2	-6.8	-6.5	6.83	-	2.124	3.476	2.80
Mean	386	369	257		957	9.20	8.82		-	0.967	2.237	
LSD at Co (A)	0.17				0.6				0.22			
LSD water (B)	1.0				0.12				0.253			
LSD (AxB)	0.34				0.072				0.256			

Table 4: Effect of cobalt on some chemical constituents of bean pods under different irrigated water levels.

Chemical constituents (%)	Total proteins (%)				Total carbohydrates (%)				Total soluble solids (%)				Vitamin "C" (mg per 100g fresh weight)			
	Irrigated water (%)															
Cobalt treatments (ppm)	100	80	60	Mean	100	80	60	Mean	100	80	60	Mean	100	80	60	Mean
Control	6.19	6.13	6.08	6.13	10.0	8.9	8.12	9.01	18.7	17.2	16.9	17.6	14.02	14.0	12.90	13.64
4	9.63	9.81	10.06	9.83	10.7	10.0	8.64	9.40	20.1	19.0	18.8	19.3	14.30	14.26	13.11	13.89
8	13.06	13.4	13.63	13.0	11.3	9.12	9.05	9.82	20.9	19.6	19.3	19.9	14.98	14.89	13.56	14.48
12	20.13	20.38	20.88	20.5	11.9	9.34	9.22	10.2	21.4	20.3	19.8	20.5	15.19	15.14	14.20	14.84
16	18.9	19.31	19.63	19.3	11.5	9.27	9.08	9.95	21.2	20.0	19.2	12.1	15.02	14.92	13.91	14.62
20	17.9	18.13	18.31	18.1	10.9	9.20	8.92	9.67	20.8	20.2	18.7	19.9	14.89	14.61	13.70	14.47
Mean	14.30	14.53	14.77		11.05	9.12	8.84		20.52	19.38	18.78		14.73	14.64	13.56	
LDS (A)	0.4				1.24				0.5				0.3			
LDS (B)	0.21				1.76				1.2				1.1			
A X B	0.084				2.18				0.6				0.33			

Abscisic acid (ABA):

Data in Table (3) also show that, the ABA showed a gradual increase under water deficit conditions (80% and 60% from plant requirement). This may confirmed by Bray (2001) who found that, under water deficit the accumulation

of ABA plays on important role in increasing plant resistance.

Data in Table (3) also indicate that, as cobalt rates in plant media increased Abscisic acid contents in plants significantly increased. Increasing Abscisic acid levels stimulate stomata closure. Cobalt application reduced plant

water loss and could be used to reduce water consumption under the condition of limited water supply or desertification. These results are in harmony with those obtained by Duan and Auge (2001).

Pods chemical constituents:-

Data in Table (4) show that, the studied chemical constituent of Bean pods such as total protein, total carbohydrates, total soluble solids along with vitamin "C" significantly decreased with the reducing the rates of irrigated water treatment (100, 80 and 60%). These results are in harmony with those obtained by Zlater and Lidon (2012) they found that the chemical and physiological contents such as lipids, sugars, total soluble carbohydrates, glycolipids and phospholipids are decreased in plants under drought conditions.

Data in Table (4) reveal the promotive effect of cobalt on Bean pods chemical content compared with untreated plants. Cobalt at 12 ppm gave the highest values of total protein, total carbohydrates, total soluble solids as well as vitamin "C" under the studied water treatment. These results are in harmony with those obtained with Castro et al., (2004) who stated that treating phaseolus seeds with cobalt had a significant promotive effect on nodulation, dry weight, physiological quality, vigor, protein and elements content in phaseolus seeds.

Vitamin "C" is the major antioxidant in plant cells and involved photo protection metal and xenobiotic detoxification the cell cycle, cell wall growth and cell expansion. It act as Co-enzyme in metabolic changes and involved in photosynthesis and respiration processes (Franceschi and Tarlyn, 2002). For human, high vitamin "C" directory in take correlates with reduced gastric cancer risk (Griffiths and Lunce, 2001).

Conclusion

Under climatic changes (drought stress), cobalt increase water use efficiency and enhance plant resistance. cobalt save 20% from Bean water requirement.

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