

PRODUCTION OF ALGAL GROWTH PROMOTERS AND STUDYING THEIR EFFECTS ON MAIZE CROP

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Abstract:

Plant growth promoters were estimated in three cyanobacterial strains (*Anabaena azollae* Strasburger, *Spirulina platensis* Geitler and *Nostoc muscorum* C. Agardh) in the presence of potassium nitrate, sodium chloride and tryptophan, with different concentrations. The highest concentrations of IAA and GA₃ were determined and applied to Maize that was planted in soil with EC (5.4). Cyanobacterial treatments were applied three times throughout the cultivation period of Maize. Mineral fertilization was applied according to the guidelines of the Egyptian Ministry of Agriculture, with 100% to control, and 75% to all other treatments. Final results showed that the highest yield was obtained by treating Maize with (*Spirulina platensis* with tryptophan, *Anabaena azollae* with both potassium nitrate and tryptophan) respectively. Those results were almost three times more than the control yield. It's recommended for the yield improvement; to use the selected strains, especially in sandy and saline soils.

Key words: Cyanobacterial hormones, IAA, GA₃, Maize, *Anabaena azollae*, *Spirulina platensis*, *Nostoc muscorum*, KNO₃, NaCl, Tryptophan, EC.

Introduction

Phytohormones are found not only in higher plants, but also in algae, and in plant-associated bacteria and fungi (Lu and Xu, 2015). Indole acetic acid (IAA) is a natural auxin that is synthesized in many species of non-seedling plants, many bacteria, fungi and algae (Varalakshmi and Malliga, 2012). Gibberellins (GAs) are tetracyclic diterpenes that occur naturally in higher plants. There is continued interest in the biosynthetic origin of the GAs since some of them are known to act as native regulators controlling a range of growth responses, including seed germination, floral development, and shoot elongation (Graebe, 1987).

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Spirulina platensis is a filamentous, photosynthetic cyanobacterium, extensively grown for human food supplements and animal feed (**Mustafa *et al.*, 1994**). The great nutritional value of *Spirulina* is due to its high protein content, polysaccharides, vitamins, and minerals (**Colla *et al.*, 2007**). It is known to produce intracellular and extracellular metabolites with diverse biological activities such as antifungal (**MacMillan *et al.*, 2002**), antiviral (**Hayashi and Hayashi, 1996**) and antibacterial activities (**Kumar *et al.*, 2011**).

The growth promotion in response to application of *Nostoc muscorum* (N-fixing cyanobacteria) could be attributed to the nitrogenous as well as nitrate reductase activities of the algae applied to the surface of plants, or the amino acids and peptides produced in cyanobacterial filtrate and other compounds that stimulated growth of plants (**Adam, 1999**).

The symbiotically associated cyanobacterium (*Anabaena azollae*) was isolated from *Azolla pinnata* and cultured under a free-living state. The strain showed high efficacy in suppressing the fungal mycelial growth of either *Fusarium oxysporum* or *Alternaria alternata* (**Abd El Aal, 2013**).

Salt stress is becoming a serious environmental issue because 7% of the land, 20% of the arable land and 50% of irrigated land in the world are salinized. The agricultural production was reported to be dramatically reduced in approximate 33% by soil salinization (**Gao *et al.*, 2011**). **Priyanka *et al.* (2016)** reported that cyanobacteria have been reported to grow extensively on both saline and alkaline soils that commonly called as "Usar". They also increase the water-holding capacity through their jelly structure, increase in soil biomass after their death and decomposition and preventing weeds growth (**Ibrahim *et al.*, 2016**).

Maize (*Zea mays* L.) is believed to have originated in central Mexico 7000 years ago from a wild grass; And Native Americans transformed Maize into a better source of food (**Smith, 2001**). Maize contains approximately 72% starch, 10% protein, and 4% fat, supplying an energy density of 365 Kcal/100 g and is grown throughout the world, with the United States, China, and Brazil being the top three Maize-producing countries in the world, producing approximately 563 of the 717 million metric tons/year (**Nuss and tanumihardjo, 2010**). Studies on Maize, involving introducing sodium chloride (NaCl) with different

concentrations have been made before and indicated that maize seed germination and chlorophyll content are sensitive to salt stress (**Aliu et al., 2015**).

The aim of this work is to study the application of cyanobacterial strains, producing IAA and GA₃, and studying their leverage on Maize crop and improving the yield both qualitatively and quantitatively, under the influence of different additives, added to the cyanobacterial media.

Materials and Methods

Microorganisms used:

Three strains of algae; *Nostoc muscorum* C. Agardh, *Anabaena azollae* Strasburger and *Spirulina platensis* Geitler were obtained from Agricultural Research Centre- Soil, Water and Environment Research Institute, Giza, Egypt (ARC,SWERI).

Production of algal growth promoters:

Media used: Both of *Nostoc muscorum* and *Anabaena azollae* were cultivated on nitrate- free medium (Blue Green “BG”11 medium) according to **Rippka et al., 1979**, while *Spirulina platensis* was cultivated on Zarrouk medium according to **Zarrouk, 1966**.

Laboratory experiment: All of three tested cyanobacterial strains were grown individually on either tryptophan or potassium nitrate (KNO₃) at concentrations: 0, 50, 100, 150 and 200 part per million (ppm) while the concentrations of NaCl were 0, 50, 100, 150 and 200 mmol. Both IAA and GA₃ were measured calorimetrically every 10 days to obtain the maximum concentrations. IAA and GA₃ were determined by Salkowski reagent according to **Glickmann and Dessaux, 1995** and Folin reagent according to **Udagwa and Kinoshita, 1961**, respectively.

Treatments could be summarized as follow:

Treatment control (Tc)= Control.

Treatment 1 (T1)=*Spirulina platensis* X NaCl.

Treatment 2 (T2)=*Anabaena azollae* X NaCl.

Treatment 3 (T3)=*Nostoc muscorum* X NaCl.

Treatment 4 (T4)=*Spirulina platensis* X KNO₃.

Treatment 5 (T5)=*Anabaena azollae* X KNO₃.

Treatment 6 (T6)=*Nostoc muscorum* X KNO₃.

Treatment 7 (T7)=*Spirulina platensis* X tryptophan.

Treatment 8 (T8)=*Anabaena azollae* X tryptophan.

Treatment 9 (T9)=*Nostoc muscorum* X tryptophan.

Field Experiment on Maize:

Before starting the experiment; complete analysis (physical, chemical and biological analyses) for the initial soil was estimated.

The seeds of *Zea mays* L. (single white hybrid 2031) were soaked in the three cyanobacterial tested strains, with the three additives, which had the highest concentrations of IAA individually for 2 hrs., then cultivated under greenhouse conditions at the ARC, SWERI. After 30 days, the same treatment was added to the soil. The three cyanobacterial tested strains that had the highest concentrations of GA₃ were added individually after 60 days of agriculture. Cyanobacterial treatments were added by 50 L/Fed (**Reddy *et al.*, 1986**). Mineral fertilization with Nitrogen, Phosphorus, and Potassium (NPK) and micronutrients was applied to Maize plants according to the standard guidelines of the Egyptian Ministry of Agriculture. The nitrogen fertilizer requirements were substituted by 75% of recommended dose throughout cyanobacterial treatments. **Mineral fertilizers:** Ammonium Nitrate was added by 400 kg/ Fed in three equal batches at preparation of soil, after 24 and 45 days of agriculture. Super Calcium

Phosphate (15%) was also added on the soil preparation with dose 200 kg/ Fed. Potassium Sulphate (48%) was added on preparation as 50 kg/ Fed. Foliar application of micronutrients was applied after 28 days of cultivation.

Soil Analyses:

Soil chemical analyses were determined after 60 days for all soil samples versus control and initial soils. Soil biological activity was evaluated in terms of total microbial counts (**Allen 1959**) and total counts of cyanobacteria (**Allen and Stanier, 1968**). Carbon dioxide (CO₂) evolution was determined according to **Gaur et al. (1971)**, Dehydrogenase activity (DHA) was estimated according to **Casida et al. (1964)**, and total Nitrogen was determined by Kjeldahl method and calculated according to **A.O.A.C. (2005)**.

Plant Analyses:

Shoot length and root weight were determined after 60 days of agriculture. Pigments such as chlorophyll a, chlorophyll b and carotenoids in ear leaves were determined according to **Metzner et al. (1965)**. Determination of NPK was observed according to **Bremner, (1965)**, and **Herbert et al., (1971)** respectively.

Total yield, weight of cobs, weight of kernels per cob and weight of 100 kernels were determined. Percentage of fixed oil in caryopses was determined according to **Horwitz, 1965**.

Results and Discussion

Determination of algal growth promoters:

As shown in table (1); it was observed that the three algal strains showed the highest IAA production values as follows:

Spirulina platensis showed the highest IAA production level (34.6 µg/ml) at concentration (conc.) 0 mmol NaCl after 10 days (control), then (34.23 µg/ml)

at conc. 200 ppm tryptophan after 20 days, and then (22.7 µg/ml) at conc. 150 ppm KNO₃ after 30 days, respectively.

Anabaena azollae showed the maximum production of IAA (31.1 µg/ml) at conc. 200 ppm tryptophan after 20 days, then (13.4 µg/ml) at conc. 100 mmol NaCl after 10 days, and finally with a very slight difference (13.2 µg/ml) at conc. 100 ppm KNO₃ after 40 days, respectively.

Nostoc muscorum could show maximum IAA production (32.5 µg/ml) at conc. 150 ppm tryptophan after 20 days, then (15.55 µg/ml) at conc. 0 mmol NaCl after 20 days (control), and then (13.2 µg/ml) at conc. 50 ppm KNO₃ after 30 days, respectively. Those results agree with (Jaiswal *et al*, 2018) who stated that IAA production in cyanobacteria (e.g. *N. muscorum* and *A. variabilis*) increases, to its maximum level, with the addition of tryptophan. Moreover, (Bhosale *et al.*, 2016), stated that IAA production is highly affected with nitrate and sodium chloride concentrations, if added to algal media.

As for GA₃ production; it was observed that:

S. platensis showed the highest conc. of GA₃ (105.25 µg/ml) at conc. 100 mmol NaCl after 10 days, then (48.23 µg/ml) at conc. 200 ppm tryptophan after 20 days, and finally (13.2 µg/ml) at conc. 200 ppm KNO₃ after 40 days, respectively.

GA₃ showed the highest levels in *A. azollae* (102 µg/ml) at conc. 200 mmol NaCl after 10 days, then (84.2 µg/ml) at conc. 100 ppm tryptophan after 20 days, and finally (12.11 µg/ml) at conc. 0 ppm KNO₃ after 20 days (control), respectively.

N. muscorum showed the maximum GA₃ (138 µg/ml) at conc. 200 mmol NaCl after 30 days, then (101 µg/ml) at conc. 100 ppm tryptophan after 20 days, and then (16 µg/ml) at 0 ppm KNO₃ after 20 days (control), respectively. It's not surprising that the highest GA₃ levels were obtained at high levels of NaCl for all algal species, as GA₃ is considered to be one of the well-known anti-stress factors identified by plants (Niu, 2014).

Table (1): The highest values of IAA and GA₃, resulted in propagation of *S. platensis*, *N. muscorum* and *A. azollae*, on different conc. of NaCl, KNO₃ and tryptophan throughout 40 days.

Treatment C	Algal Strain	Additive	Concentration	Time (days)	Concentration (µg/ ml)	
					IAA	GA ₃
T1	<i>Spirulina platensis</i>	NaCl	0	10	34.6	105.25
		NaCl	100 mmol	10		
T2	<i>Anabaena azollae</i>	NaCl	100 mmol	10	13.4	102
		NaCl	200 mmol	10		
T3	<i>Nostoc muscorum</i>	NaCl	0	20	15.55	138
		NaCl	200 mmol	30		
T4	<i>Spirulina platensis</i>	KNO ₃	150 ppm	30	22.7	13.2
		KNO ₃	200 ppm	20		
T5	<i>Anabaena azollae</i>	KNO ₃	50 ppm	30	13	12.11
		KNO ₃	0	20		
T6	<i>Nostoc muscorum</i>	KNO ₃	50 ppm	30	13.2	16
		KNO ₃	0	20		
T7	<i>Spirulina platensis</i>	Tryptophan	200 ppm	20	34.23	48.23
		Tryptophan	200 ppm	20		
T8	<i>Anabaena azollae</i>	Tryptophan	200 ppm	20	31.1	84.2
		Tryptophan	100 ppm	20		
T9	<i>Nostoc muscorum</i>	Tryptophan	150 ppm	20	32.5	101
		Tryptophan	100 ppm	20		

Field experiments:

Initial soil analysis; as shown in table (2), showed that the initial EC was 5.4 mS/ cm, and pH 7.6 with sandy texture. The initial total N was found to be 53.375 (mg/ Kg), CO₂ evolution was found to be 329 (mg CO₂/ 100 gm soil), Dehydrogenase activity was estimated and found to be 54.9 (µgTPFg-1 dry soil day-1), Mean Count Cyanobacteria was 2.9 (10³ cfu/ gm soil) while Mean Count Bacteria was 49.6 (10⁶ cfu/ gm soil).

Table (2): Physical , chemical and biological analyses of Initial Soil

Physical analysis of soil										
Coarse sand	Fine sand	silt	Clay				Texture			
34%	61.40%	1.60%	3.06%				Sandy			
Chemical analysis of soil										
Anions meq / L			Cations meq / L							
SO ₄ ⁻²	Cl ⁻	HCO ₃ ⁻	K ⁺	Mg ⁺²	Na ⁺	Ca ⁺²	EC	pH	Total N (mg/Kg)	SP (%)
41.19	22.88	2.83	0.97	11.56	8.32	46.05	5.4 mS/cm	7.6	53.38	28
Biological analysis of soil										
CO ₂ Evolution		Mean bacterial count	Mean cyanobacterial count			Dehydrogenase activity				
329 (mg CO ₂ / 100 gm soil)		49.6 (10 ⁶ cfu/ gm soil)	2.9(10 ³ cfu/ gm soil)			54.9 (µg TPF g/ dry soil/ day)				

Cultivation of Maize with different cyanobacterial treatments, using the highest IAA and GA₃ conc.; showed a remarkable decrease in the soil EC in most treatments, as shown in table (3), compared to the initial soil EC. This agrees with (Noufal *et al*, 2012) who stated that soil EC is significantly decreases with addition of bio-fertilizers to the soil. pH showed an elevation due to addition of the different additives, however, it did not affect the microbial activity, which was determined with CO₂ evolution, Dehydrogenase activity, Total count of both bacteria and cyanobacteria. Moreover, it was generally observed that the highest characters of the field experiment took place around the pH 8 and 8.1; this may be resulted from the optimum rate of IAA production at such pH, as explained by (Bhosale *et al.*, 2016).

Table (3): Soil chemical analyses after 60 days

Treatment	EC (mS/ cm)	pH	Total N (mg/ Kg)
Tc	2.7	7.8	55.51
T1	1.28	7.9	59.78
T2	1.42	7.9	85.4
T3	0.75	7.9	81.13
T4	1.14	8.1	68.32
T5	1.37	8	57.65
T6	1.23	8.1	59.78
T7	3.42	8.1	89.67
T8	2.77	8	122.7
T9	3.7	8	153.72

Total N showed a remarkable elevation on addition of cyanobacterial treatments. The highest N % was reported at the treatment T9 (*Nostoc muscorum* with tryptophan) with 153.72 mg/Kg, compared to control (55.51 mg/ Kg). This result agrees with (**Aziz *et al.*, 2014**) who stated that the beneficial effect of algalization to crop yield components could be attributed to the ability of cyanobacteria in N₂-fixation as well as producing the growth promoting substances such as ascorbic acid, auxins and vitamin B₁₂.

As for the soil microbial analyses; the algal treatments had remarkable effects on the results. As shown in figure (1); CO₂ evolution showed the highest levels at treatment T6 (*Nostoc muscorum* with KNO₃) with 278.7mg CO₂/ 100 gm soil, compared to control (85.8). Dehydrogenase activity showed the highest level at the treatment T7 (*Spirulina platensis* with tryptophan) with 105.1 (µgTPPg-1 dry soil day-1), compared to control (29.23), as shown in figure (2). Mean Cyanobacterial Count showed the maximum level ; as shown in figure (3); at the treatment T4 (*Spirulina platensis* with KNO₃) with 19.7(10³cfu/ gm soil), compared to control (4.85), while Mean Bacterial Count showed maximum determination; as shown in figure (4); with treatment T8 (*Anabaena azollae* with tryptophan) with 97(10⁶cfu/ gm soil), compared to control (79).

Table (4) showed the plant analyses after 60 days. Maximum shoot height of Maize was determined with treatment T8 (*Anabaena azollae* with tryptophan) with 271 cm, compared to control (224.2 cm), this agrees with (**Mohan and Kumar, 2017**) who reported maximum shoot length on applying different algal species to several field crops. Maximum dry weight of root was obtained at treatment no. T6 (*Nostoc muscorum* with KNO₃) with 17.85 gm, compared to control (16.62) gm, however; other algal treatment showed higher percentage of root hairs. This explains the relatively lower weights of other specimens. This agrees with (**Spaepen *et al.*, 2007**) who explained that at moderate levels of IAA, microbes use 1 Aminoacetylpropane-1 Carboxylate Synthase (ACC) (Ethylene precursor) as a nitrogen source, thus limiting the ethylene production and enhance root elongation.

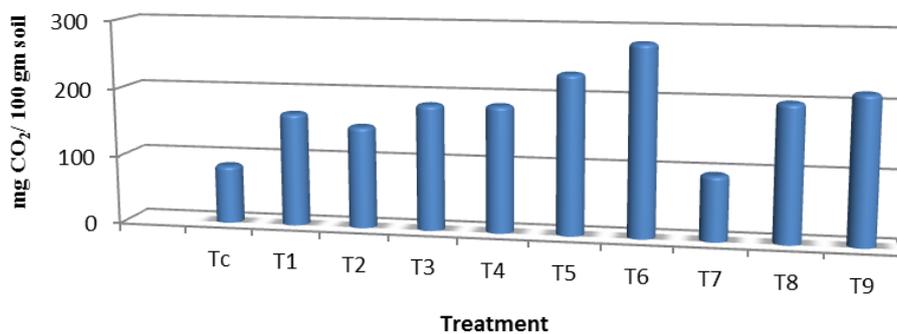


Figure (1): CO₂ Evolution in soil after 60 days (mg CO₂/100 gm soil)

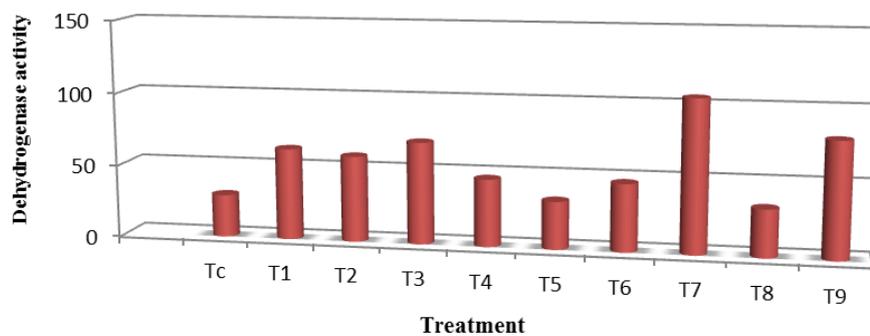


Figure (2): Dehydrogenase activity in soil after 60 days (μgTPFg^{-1} dry soil day⁻¹)

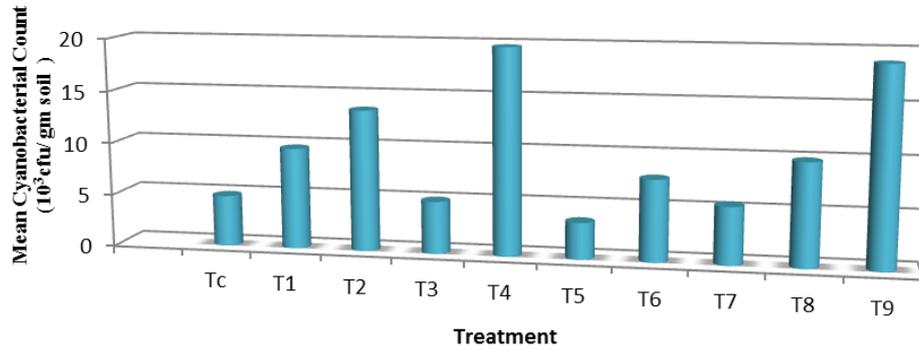


Figure (3): Mean Cyanobacterial Count in soil after 60 days (10³cfu/ gm soil)

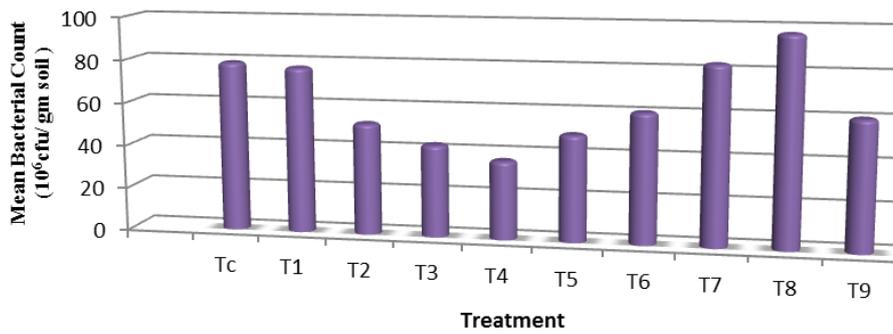


Figure (4): Mean Bacterial Count in soil after 60 days (10⁶cfu/ gm soil)

Table (4): plant analyses after 60 days

Treatment	Shoot Height "cm"	Root Dry weight "gm"	Chlorophyll a ($\mu\text{g}/\text{ml}$)	Chlorophyll b ($\mu\text{g}/\text{ml}$)	Carotenoids ($\mu\text{g}/\text{ml}$)
Tc	224.2	16.62	19.94	10.48	2.36
T1	215	10.12	21.01	8.56	3.78
T2	180.2	16.98	11.46	3.2	2.54
T3	210	6.79	21.88	9.73	3.75
T4	233.2	14.54	22.1	10.67	3.33
T5	235	8.29	21.23	9.33	3.72
T6	227.2	17.85	21.32	9.86	3.5
T7	258.6	6.34	22.84	14.51	2.83
T8	271	9.68	21.26	9.24	3.84
T9	240.4	16.71	22.71	11.51	3.37
Least Significant Difference (L.S.D.) at 0.05%	7	0.97	1.77	2.3	0.383

Chlorophyll a showed maximum level at the treatment T7 (*Spirulina platensis* with tryptophan) with 22.84($\mu\text{g}/\text{ml}$) compared to control (19.94 $\mu\text{g}/\text{ml}$). Chlorophyll b showed the highest level also at the treatment T7 (*Spirulina platensis* with tryptophan) and with 14.5($\mu\text{g}/\text{ml}$) compared to control (10.48 $\mu\text{g}/\text{ml}$) respectively. The highest carotenoids level was obtained with T8 (*Anabaena*

azollae with Tryptophan) with (3.84µg/ ml) compared to control (2.4µg/ ml). Those results are in correspondence with **(Aziz *et al.*, 2014)**.

As for NPK analysis, it was observed that there were tremendous differences in N absorption in most cases compared to control. As shown in table (5); the highest N percentage was obtained by T9 (*Nostoc muscorum* with tryptophan) with rate (1.9%) compared to control (0.5%); those results agree with **(Mohan *et al.*, 2015)**. P estimation also showed bigger absorption rates than that of control. Most increment was estimated by T2 (*Anabaena azollae* with NaCl) with (0.22%), compared to control (0.16%). K absorption did not show a real change at the 60 days period, however, it was highly more affected at the harvest period.

At the harvest; the yield of Maize, treated with the selected algal treatments showed real differences in total yield production, mean weight of cobs per plant, weight of kernels per cob, and weight of 100 kernels, compared to control. Generally, EC highly affected the control; this agrees with **(Bilgin *et al.*, 2008)**, who stated that Maize is negatively affected starting EC 2.7.

In table (6); NPK at harvest was estimated. The highest percentage of N absorption was reported for treatment T7 (*Spirulina platensis* with tryptophan) with 1.4%, compared to control (0.65%). P showed the maximum absorption rate at T7 as well with (0.43%), compared to control (0.32%). The highest K absorption rate was reported to T2 (*Anabaena azollae* with NaCl) with 1.4%, compared to control (1.2%).

Table (5): Plant NPK analysis after 60 days

Treatment	N%	P%	K%
Tc	0.5	0.16	1.42
T1	0.4	0.18	1.08
T2	0.4	0.22	1.33
T3	0.2	0.12	0.98
T4	1.69	0.13	1.12
T5	0.65	0.12	1.35
T6	0.9	0.17	1.15
T7	0.9	0.14	1.31
T8	1	0.16	1.26
T9	1.9	0.2	1.17
L.S.D. at 0.05%	0.15	0.03	0.26

Table (6): Plant NPK analyses at harvest

Treatment	N%	P%	K%
Tc	0.65	0.32	1.2
T1	0.9	0.25	1.33
T2	1.25	0.29	1.4
T3	1	0.12	1.24
T4	0.65	0.22	0.71
T5	0.65	0.28	1.01
T6	1.15	0.22	1.1
T7	1.4	0.43	1.17
T8	0.2	0.27	1.012
T9	0.3	0.14	1.24
L.S.D. at 0.05%	0.12	0.03	0.16

As shown in figure (5); the highest yield was obtained with treatment T7 (*Spirulina platensis* with tryptophan) with 23.22 ardeb/ Fed., compared to control (7.5 ardeb/ Fed). This agrees with (Wu *et al*, 2017) who stated that salinity could decline the crop yield up to 60 %.

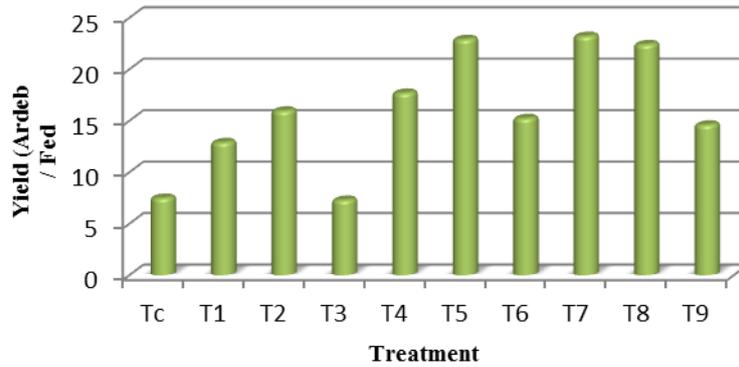


Figure (5): Yield Analysis at Harvest (Ardeb/ Fed)

Table (7) showed that the highest mean weight of cobs was also obtained with T7 (*Spirulina platensis* with tryptophan) by 259.2 gm, compared to control (123.94 gm). The highest mean weight of kernels was observed by T7 (*Spirulina platensis* with tryptophan) with a value of 183.03 gm, compared to control (76.4 gm). The treatment T9 (*Nostoc muscorum* with tryptophan) had the highest weight of 100 kernels with value 40.62 gm, compared to control (33.33 gm).

As for calculating the percentage of fixed oil (Figure (6)), it was found that the highest % was reported for treatment T7 (*Spirulina platensis* with tryptophan) with 14.88%, compared to control (13.56), with least significant difference 0.8.

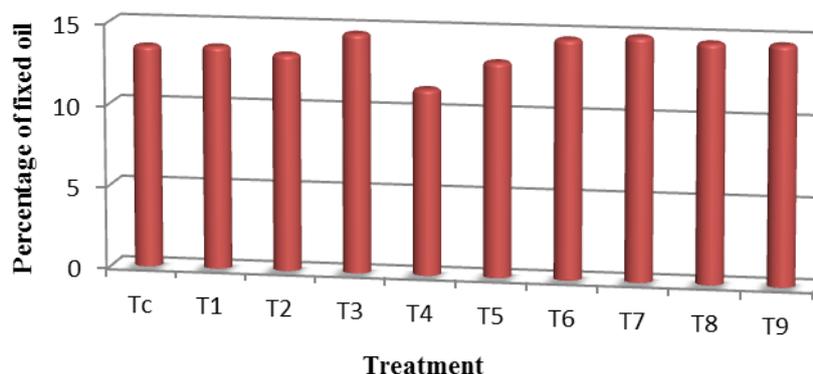


Figure (6): Percentage of Fixed Oil at Harvest

Table (7): Plant analyses at harvest

Treatment	Cobs Wt. (gm)	Kernels Wt. (per cob) (gm)	Dry wt. of 100 kernels
Tc	123.94	76.4	33.33
T1	157.6	110.34	38.86
T2	168.03	114.6	31.72
T3	77.28	50.2	31.66
T4	101.78	65.13	30.25
T5	180.11	133.42	38.34
T6	191.84	135.1	36.47
T7	259.2	183.03	38.18
T8	209.92	143.79	39.55
T9	181.88	127.624	40.62
L.S.D. at 0.05%	47	42	---

Conclusion

Applying different cyanobacterial strains, treated with different additives, to Maize that was cultivated in a soil affected with relatively high EC, was found to overcome such stress, if applied during the optimum time of IAA and GA₃ production by cyanobacteria. It's recommended for the yield improvement; to use the selected strains, especially in sandy and saline soils, as it was found that using treatments T7 (*Spirulina platensis* with tryptophan), T5 (*Anabaena azollae* with KNO₃) and T8 (*Anabaena azollae* with tryptophan), each separately; can improve the yield three times more than control. while treatments T2 (*Anabaena azollae* with NaCl), T4 (*Spirulina platensis* with KNO₃), T6 (*Nostoc muscorum* with KNO₃) and T9 (*Nostoc muscorum* with tryptophan) could improve the yield two times more than control.

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إنتاج محفزات النمو من الطحالب ودراسة تأثيرها علي محصول الذرة

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تم تعيين محفزات النمو النباتية في ثلاث سلالات من السيانوبكتيريا (*Spirulina platensis* Geitler) و (*Anabaena azollae* Strasburger و *Nostoc muscorum* C. Agardh) ، وذلك في وجود كل من نترات البوتاسيوم، كلوريد الصوديوم، والتريبتوفان، بعدة تركيزات مختلفة. تم تحديد أعلى نسب لكل من الإندول والجبريلين، وتم عمل تجربة حقلية علي محصول الذرة، والذي تمت زراعته بمركز البحوث الزراعية، في تربة ذات توصيل كهربي 5.4. تم تطبيق المعاملات الطحلبية ثلاث مرات خلال دورة زراعة المحصول. تمت إضافة الأسمدة المعدنية طبقاً لإرشادات وزارة الزراعة المصرية، مع تطبيق نسبة 100% من السماد للكنترول، وال75% لباقي المعاملات. أوضحت النتائج النهائية أن أعلى محصول تم تقديره في معاملة محصول الذرة ب *Spirulina platensis* مع التريبتوفان، و *Anabaena azollae* مع نترات البوتاسيوم، ثم *A. azollae* مع التريبتوفان. أظهرت النتائج ان المعاملات المذكورة أعطت إنتاجية أكبر من الكنترول بحوالي ثلاث مرات، ومن ثم يوصي باستخدام هذه المعاملات، لا سيما في الأراضي الرملية، أو الأرضية المتأثرة بالملوحة.