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Research Article

Effect of Water Stress and Biological Fertilization on Maize Growth, Chemical Composition and Productivity in Calcareous Soil

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Abstract

Objective: The present study was to estimated maize growth, chemical composition and productivity in calcareous soil under the effect of bio-fertilizer. **Methodology:** Two field experiments were conducted at Department of Botany, Faculty of Science, Zagazig University to study the effect of two different bio-fertilization treatments (*Azotobacter crococcum*+*Mycorrhiza*+*Pseudomonas* spp.) and (*Azospirilium lipoferum*+*Bacillus megatherium* var. *phosphaticum*+*Bacillus subtillus*) accompanied with two mineral NPK levels {full (100%) and half (50%) dosages}, on maize cv. 30B9, under four water regime treatments; which missed one irrigation (second, third and fourth) beside the normal irrigation treatment as a control. **Results:** The obtained results confirmed that maize plants can tolerate water scarcity at the vegetative growth period (60 days). Missing the fourth followed by the third irrigation seemed to be the best after the control one. The bio-fertilization treatments, helped the plants to overcome the negative effects of water stress at any growth period. The first bio-fertilizer appeared to be more effective under water stress conditions if compared with the second treatment. **Conclusion:** Moreover, the full dosage of the mineral NPK fertilizer was found be the best followed by both first and the second bio fertilizer accompanied with the half dosage of the mineral NPK. It could be mentioned that, using the half dosage of the mineral NPK, the bio-fertilizers will be more logic.

Key words: Maize, water stress, biological fertilization, calcareous soil

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Competing Interest: The authors have declared that no competing interest exists.

Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

Maize plant considered as one of the most important goals of the Egyptian government in order to face the human and animal essential needs. In this respect, to increase the maize production, a continuous extension efforts had been done at both horizontal and vertical levels. Kang *et al.*[1] stated that the climatic condition all over the world was changed under the influence of global warming. This will create unusual weather phenomena often in the form of water deficit or floods and water logging. The plants should be adapted to face such stress. This adaptation came from genetic machineries which aid plant to produce enzymes, proteins and synthesize molecules suitable to combat water shortage. Denmead and Shaw[2], Norwood and Dumler[3], Derby *et al.*[4], Barnaby *et al.*[5], Perdomo *et al.*[6] and Velikanov *et al.*[7] concluded that maize plant is considered as a very sensitive crop to water stress especially at reproductive phase. Plants can tolerate the water stress in the vegetative growth period if compared with the other growth periods. The total sensitive period to water stress was found to be at the least 55 days of plant growth. Reaid *et al.*[8] and Andraski and Bundy[9] claimed that the scarcity in irrigation water associated with inapplicable fertilization treatments and the pollution made by the fertilizers itself particularly under new reclaimed soil conditions represent a great challenges faced all efforts made in this target. Water stress has an important effect on water consumption and maize yield. A positive linear relationship between grain yield and water use has been recognized by several authors[10,11]. It was found that water stress reduce plant height, decrease leaf area index[12], this resulting in a decrease in the rate of photosynthesis and contributes to a reduction in biomass yield. It was found that dry matter production of non-stressed plants is usually high compared to stressed plants, this is because water stressed plants cannot use solar radiation effectivelly[13]. Moreover, Nour El-Din *et al.*[14], Yakout *et al.*[15], Reaid *et al.*[8], Zhang *et al.*[16], Zhao *et al.*[17] and Liu *et al.*[18] stated that maize plant is considered as a greedily plant to fertilization, particularly to nitrogen when irrigation water is available but when there is a scarcity in irrigation water, fertilization is not an acceptable risk. Therefore, biological fertilizers may supply maize plant with all nutrients needed for plant metabolism and growth without all hazards occurred when applying chemical fertilizers under water stress conditions Koliai *et al.*[19]. Many reports declared the associations of N_2 fixing bacteria, phosphate bacteria and Mycorrhiza with plant root system[20-22]. Moreover, Edmeades *et al.*[23] observed that water stress significantly decreases number of seeds per row. Also, they mentioned micro-organisms in bio- fertilizers are to increase absorption of food elements[24], they work to solve insoluble phosphate by production of organic acids that obtained as exo-acids from sugars through reaction in the rhizosphere and this absorption of elements result in crops with increased yield[25-27]. Schweiger and Jakobsen[28] and Mozafar *et al.*[29] reported that, under water stress, the bio-fertilizers helped the plants to overcome the negative effects of water stress and increased significantly all growth characters, chemical composition and consequently yield and its attributes. Moreover, Sundara *et al.*[30] found that *Bacillus megatherium* var. *phophaticum* increased nutrient availability especially phosphorus in the soil. It also enhanced sugar cane growth, yield and quality. Sharma and Singh[31], Yuan *et al.*[32] and Cherian *et al.*[33] showed that mixed application of chemical and biological fertilizers influenced the physiology of plants through increasing amounts of photosynthetic substances, which served to change the flow of photosynthetic substances to stems, roots and affected the absorption of soil minerals. This matter often changes the nutritional condition of host plant tissue that facilitates better absorption and a higher rate of photosynthesis as a result, increases evaluations for the trait of plant dry weight under water stress conditions. Amanolahi-Baharvand *et al.*[34], Lazcano *et al.*[35] and Hoflich *et al.*[36] claimed that the useful effect of integrating microbial fertilizer with chemical fertilizer to increase seed yield under water stress conditions.

The objective of this study is to estimate maize growth, chemical composition and productivity under the effect of two bio-fertilizers (NPK) associated with either 0, 50 or 100% of chemical NPK fertilizers under different water regime treatments during the period of vegetative growth.

MATERIALS AND METHODS

Two field experiments were conducted in summer seasons at Department of Botany, Faculty of Science, Zagazig University to investigate the response of maize plant (*Zea mays* L. var. *pioneer* 30B9) to different bio-fertilization treatments (*Azotobacter crococcum*+*Mycorrhiza* +*Pseudomonas* spp.) and (*Azospirilium lipoferum*+*Bacillus megatherium* var. *phosphaticum*+*Bacillus subtillus*) accompanied with two mineral NPK levels {full (100%) and half (50%) dosages}, under four water regime treatments which included missed one irrigation treatment after El-mohayah irrigation 10 days post germination, the 2nd irrigation (25 days after germination), the 3rd irrigation (40 days after germination) and the 4th irrigation (55 days after germination) beside the normal irrigation treatment as a control. Inoculants used consisted of three strains of every microorganism to protect from bacteriophage in the rhizosphere. The inoculated maize plant received 3 mL of inoculum/hill just before the first irrigation (El-mohayah). Bacterial cultures used for inoculates normally had a cell density of 10 with 10^6/plant, where Mycorrhiza was 60 spore/plant. They were grown in a liquid medium containing 1 g NH_4Cl/liter[37]. Consequently, spores of Mycorrhiza were isolated from the soil by the wet sieving and decanting method[38].

Compost (complete fermented organic materials) was added into the soil during soil preparation in the dosage 20 kg/fed accompanied with phosphorus as calcium super phosphate P_20_5 in the rate 15 and 30 kg P_20_5/ fed following the treatments scheme. The other chemical fertilizers were added as complete and half of the recommended dosage i.e., nitrogen as ammonium nitrate 3 3.5% N in the rate 60 and 120 kg N/fed., while potassium was added as potassium sulfate 40% K_2SO_4 in the rate 12 and 24 kg K_2O_4/fed in two equal dosage added just before the first and second irrigation.

Split split plot design in four replicates was used in this experiment, where irrigation treatments occupied the main plots, chemical fertilizers in the sub-main and bio-fertilizers in the sub-sub main plots. The experimental plot area was 14.20 m^2 with four rows of 4 m in length and 71 cm in width.Two grains per hill were sown in the first of June in both seasons at distance of 20 cm. Plants were thinned after 25 days from sowing to one plant/hill. Samples were taken one week after applying the fourth irrigation for studying some growth characters i.e., plant height (cm/plant), number of leaves/ plant, fresh and dry weights (g/plant), third leaf area (cm^2) (using "Li-3000A" portable leaf area meter) and chemical composition i.e., total nitrogen percentage according to the method described by Paech and Tracey[39], potassium percentage according to Johnson and Ulrich[40], phosphorus percentage following the method described by John[41] and total pigments using SPDA-502 leaf chlorophyll meter, then converted into total chlorophyll (a+b) referring to the equation published by Markwell *et al*.[42]. Similarly, yield and its components were evaluated at harvest time i.e., ear weight' cm, number of grains/ear, seed index (as 100 grain weight/g), biological and grain yield (t/fed). Data obtained during both seasons was exposed to the proper method of statistical analysis of variance (ANOVA) as described by Steal and Torrie[43] and Duncan's new multiple range test was used to differentiate between means as described by Duncan[44].

RESULTS

Results in Table 1 indicated that there were significant differences between irrigation treatments i.e., it was found that normal irrigation treatment (control) appeared to be the best irrigation treatment since it enhanced all growth characters (plant height, fresh, dry weights, number of leaves and leaf area), followed by the other treatments including (missing either the second, the third or the fourth irrigation),

Table 1: Response of some characters of maize plants to irrigation treatments

Studied characters	Normal irrigation	Missing second irrigation	Missing third irrigation	Missing forth irrigation
Growth characters				
Plant height/cm	238^A	210.7^B	195.1^C	158.1^D
No. of leaves/plant	15.6^A	13.3^B	12^C	10.4^D
Fresh weight g/plant	843.2^A	606.4^B	508^C	386.3^D
Dry weight g/plant	156.6^A	110.4^B	88^C	63.2^D
Leaf area/cm^2	508.7^A	433.2^B	379^C	297.3^D
Chemical composition				
Pigment	51.4^A	45.7^B	42.1^C	35.8^D
Chlorofyll (μmol m^{-1})	739.5^A	629^B	532.4^C	414.4^D
N (%)	1.72^A	1.44^B	1.23^C	1.05^D
P (%)	0.94^A	0.72^B	0.61^C	0.52^D
K (%)	2.87^A	2.44^B	2.35^C	1.97^D
Yield and its components				
Ear weight/g	340.1^A	324.6^B	240.9^C	176.8^D
No. of grains/ear	529.7^A	447.5^B	842.2^C	631.0^D
Bio yield (t/fed)	8.04^A	6.48^B	5.28^C	3.98^D
Grain yield (t/fed)	3.26^A	2.76^B	2.28^C	1.78^D
100 Grain weight	25.83^A	21.13^B	92.4^C	12.8^D

Mean having the same capital letters in the same row are not significantly differed at $p \geq 0.05$

respectively. Similarly, Table 1 showed that water stress reduced significantly the absorption of nitrogen; phosphorus and potassium by plant, also total pigments and total chlorophyll accumulation were reduced. Plants under missing the fourth irrigation had the lowest growth characters and chemical composition, if compared with missing the third, the second or the normal irrigation (control). These results clarified that the unfavorable obtained growth characters was due to the low plant nutrition. On the other hand, Table 1 indicated that, water stress affected negatively ear weight, number of grains per ear, 100 grain weight, hence, both biological and grain yield. It could be concluded that the more advanced growth stage is the more sensitive to water stress in maize plants. Since it was noticed that the second missing irrigation at 25 days produce appreciated grain yield (2.76 t fed^{-1}) compared with missing the third (2.28 t fed^{-1}) and the fourth irrigation (1.78 t fed^{-1}).

Regarding to bio-fertilization treatments results in Table 2 indicated that using the first bio-fertilizer which consist of (*Azotobacter crococcum*+Mycorrhiza+ *Pseudomonas* spp.) was more superior if compared with the second bio-fertilizer which consist of (*Azospirilium lipoferum*+ *Bacillus megatherium* var. *phosphaticum*+*Bacillus subtillus*), since the first biofertilizer enhanced all studied growth characters i.e., plant height, fresh, dry weights, number of leaves per plant and leaf area. Also, it led to enhance NPK absorption by maize plants; as a result plant total pigments and total chlorophyll content were significantly increased. Accordingly using the first biofertilizer increased significantly all studied yield components, hence biological and grain yield t/fed. With respect to NPK mineral fertilization treatment, results in Table 2 clarified that the usage of chemical NPK fertilization increased significantly all the studied growth characters, which led to significant increase in all studied plant chemical composition. This led to enhance yield components, biological and grain yield. Mineral fertilization seemed to be more applicable to get an appreciated yield if compared with bio-fertilizers application from commercial point of view, consequently highly response of maize plants to fertilization particularly to nitrogen. But when we consider the environmental costs which include the water, air and soil pollution, chemical fertilization will have high environmentally costs if compared with the bio-fertilizers application, especially when an appreciated yield is obtained and particularly, if there is no significant differences in both biological and grain yield were observed. This was noticed by applying bio-fertilizer-l as compared with 100% NPK chemical fertilizers Table 2.

Regarding to bio-fertilization treatments under different NPK Levels, data recorded in Table 3 demonstrated that Bio-fertilzer-1 seemed to be more successive under 50% of the recommended NPK dosage comparing with the Bio-fertilizer-2. Both bio-fertilizers treatments increased significantly all studied maize plants growth characters and chemical composition which significantly increased all studied yield components, biological and grain yield. When 100% of the recommended NPK chemical fertilizer dosage was applied together with bio-fertilizer 1 or 2, a significant decrease obtained in each of all studied growth characters, chemical composition, yield and its components of maize plants, respectively, if compared with the combination included 50% of mineral NPK application and either bio-fertilizer 1 or 2.

Table 2: Response of some characters of maize plant to NPK mineral fertilization and Bio-fertilization treatments

Studied characters	Control NPK 100%	Control NPK 50%	Bio-fertilizer 1	Bio-fertilizer 2
Growth characters				
Plant height/cm	202^{A}	184.6^{B}	205.1^{A}	200.7^{B}
No. of leaves/plant	13.8^{A}	11.9^{B}	12.95^{B}	12.5^{B}
Fresh weight g/plant	656.7^{A}	494.2^{B}	598.3^{B}	581.3^{C}
Dry weight g/plant	166.3^{A}	85.1^{B}	107.4^{B}	99^{C}
Leaf area/cm^2	423.8^{A}	363.8^{B}	464.1^{A}	402.2^{B}
Chemical composition				
Pigment	44.7^{A}	40.1^{B}	44.9^{A}	43.3^{B}
Chlorofyll (μmol m^{-1})	599.2^{A}	506.4^{B}	595.7^{A}	579.3^{B}
N (%)	1.37^{A}	1.2^{B}	1.4^{A}	1.38^{A}
P (%)	0.74^{A}	0.62^{B}	0.71^{B}	0.7^{B}
K (%)	2.92^{A}	2.86^{B}	2.39^{B}	2.39^{B}
Yield and its components				
Ear weight/g	274.4^{A}	231.9^{B}	290.4^{A}	262.4^{B}
No. of grains/ear	420.8^{A}	340.3	418.8^{A}	407.5^{B}
Bio yield (t/fed)	6.24^{A}	5.26^{B}	6.12^{A}	5.91^{B}
Grain yield (t/fed)	2.63^{A}	2.23^{B}	2.59^{A}	2.52^{B}
100 Grain weight	20.1^{A}	16.72^{B}	19.73^{B}	19.15^{B}

Mean having the same capital letters in the same row are not significantly differed at $p \geq 0.05$

Table 3: Response of some characters of maize plants to the interaction between bio fertilizer and NPK fertilization treatments

	Bio fertilizer 1		Bio fertilizer 2		Control
Treatments studied characters	NPK 50%	NPK 100%	NPK 50%	NPK 100%	NPK 100%
Growth character					
Plant height/cm	214.6^{A}	204.7^{C}	208.7^{B}	201.1^{D}	202.1^{D}
No. of leaves/plant	13.4^{A}	12.8^{B}	13.1^{A}	12.8^{B}	13.8^{A}
Fresh weight g/plant	647^{A}	597^{B}	627.5^{A}	578.1^{B}	656.7^{A}
Dry weight g/plant	115.3^{A}	107.4^{B}	110.9^{A}	103.5^{B}	116.3^{A}
Leaf area/cm^2	436.5^{A}	413.3^{B}	422.8^{A}	401.5^{B}	423.8^{A}
Chemical composition					
Pigment	46.2^{A}	44.9^{A}	45.6^{B}	44.1^{C}	44.7^{C}
Chlorofyll (μmol m^{-1})	630.1^{A}	594.8^{A}	617.9^{A}	578.4^{B}	599.2^{A}
N (%)	1.43^{A}	1.39^{B}	1.42^{A}	1.39^{B}	1.37^{B}
P (%)	0.76^{A}	0.7^{C}	0.73^{B}	0.69^{C}	0.74^{B}
K (%)	2.47^{A}	2.37^{B}	2.43^{B}	2.33^{B}	2.83^{A}
Yield and its components					
Ear weight/g	282.2^{A}	343.4^{A}	274.2^{C}	263.1^{C}	274.4^{C}
No. of grains/ear	443.8^{A}	416.3^{A}	426.3^{B}	408.7^{E}	420.8^{B}
Bio yield (t/fed)	6.55^{A}	6.11^{C}	6.32^{B}	5.78^{D}	6.24^{B}
Grain yield (t/fed)	2.71^{A}	2.58^{C}	2.63^{B}	2.52^{C}	2.63^{B}
100 grain weight	20.73^{A}	19.64^{C}	20.09^{B}	19.21^{C}	20.1^{B}

Mean having the same capital letters in the same row are not significantly differed at $p \geq 0.05$

Table 4: Response of some growth characters of maize plant to irrigation, bio-fertilization treatments

	Normal irrigation		Missing second irrigation		Missing third irrigation		Missing forth irrigation	
Studied characters	Bio 1	Bio 2	Bio 1	Bio 2	Bio 1	Bio 2	Bio 1	Bio 2
Growth characters								
Plant height/cm	240.7^{Ba}	234^{Ca}	211.7^{Bb}	209.7^{Cb}	389.4^{Ac}	381.5^{Ac}	322^{Ad}	305.7^{Bd}
No. of leaves/plant	15.6^{Ba}	15^{Ca}	13.3^{Bb}	13.1^{Bb}	91.7^{Ac}	89.3^{Ac}	71.1^{Ad}	70.3^{Bd}
Fresh weight g/plant	837.6^{Ba}	803^{Ca}	611.4^{Bb}	598.4^{Cb}	521^{Ac}	512.7^{Ac}	423.2^{Ad}	411.3^{Bd}
Dry weight g/plant	155.1^{Ba}	146.3^{Ca}	111.7^{Bb}	108.3^{Cb}	12^{Ac}	12^{Ac}	10.9^{Ad}	10.7^{Bd}
Leaf area/cm^2	487.1^{Ba}	489.5^{Ca}	436.7^{Bb}	432^{Cb}	198.3^{Ab}	196.5^{Ac}	169.6^{Ad}	162.5^{Bd}
Chemical composition								
Pigment	61.8^{Bb}	50.8^{Ca}	47.1^{Ab}	46.3^{Bb}	42.9^{Ac}	42.5^{Ac}	37.9^{Ad}	37.1^{Bd}
Ch. (μmol m^{-1})	743^{Bb}	724^{Ca}	638.4^{Bb}	620.5^{Cb}	548.7^{Ac}	539.9^{Bc}	452.5^{Ad}	432.5^{Bd}
N (%)	1.75^{Bb}	1.69^{Ca}	1.45^{Bb}	1.44^{Bb}	1.25^{Ac}	1.23^{Bc}	1.16^{Ad}	1.15^{Bd}
P (%)	0.95^{Bb}	0.92^{Bc}	0.73^{Bb}	0.71^{Cb}	0.62^{Ac}	0.61^{Ac}	0.55^{Ad}	0.54^{Bd}
K (%)	2.63^{Bb}	2.53^{Ca}	2.44^{Bb}	2.44^{Bc}	2.36^{Ac}	2.36^{Ac}	2.14^{Ad}	2.07^{Bd}
Yield and its components								
Ear weight/g	430.1^{Ba}	329.1^{Ca}	390^{Ab}	285.7^{Cb}	250.8^{Ac}	244.7^{Bc}	180^{Ad}	190.1^{Ad}
No. of grains/ear	527.3^{Ba}	512.7^{Ca}	453^{Bb}	445.2^{Cb}	400.9^{Ac}	392.1^{Bc}	294^{Ad}	280^{Bd}
Bio yield (t/fed)	8.08^{Ba}	7.78^{Ca}	6.58^{Bb}	6.38^{Cb}	5.48^{Ac}	5.28^{Bc}	4.28^{Ad}	4.08^{Ad}
Grain yield (t/fed)	3.28^{Ba}	3.18^{Ca}	2.78^{Ab}	2.78^{Ab}	2.38^{Ac}	2.38^{Ac}	1.98^{Ad}	1.78^{Ad}
100 grain weight	25.4^{Ba}	24.5^{Ca}	21.3^{Ab}	21.3^{Ab}	18.2^{Ac}	17.7^{Bc}	13.93^{Bd}	13.4^{Ad}

Mean having the same capital letters in the same row are not significantly differed at $p \geq 0.05$

When we studied the behavior of the two bio-fertilizers under different irrigation treatments, Table 4 proved that, bio-fertilizer-1 seemed to be superior when compared with bio-fertilizer-2, especially when water stress is existed. Under the severe water stress conditions as the fourth irrigation was escaped due to irrigation water deficit, bio-fertilizer I then 2 remained capable to increase all yield components in addition to biological and grain yield as a result of enhancing significantly all studied plant chemical composition and thus all studied growth characters. Similarly, when missing either 2nd or 3rd irrigation, water stress may not be as much of missing the 4th irrigation, because each of bio-fertilizer 1 then 2 could easily encourage significantly all studied growth characters as a result of increasing significantly plant chemical composition thus, yield and its attributes. When irrigation water is adequate enough to provide maize with its water requirements, although the dosage 100% of NPK is highly recommended for maize production because it is greedily to fertilizers particularly to nitrogen, but it is highly environmental costing at the same time. So, bio-fertilizer-l was superior to encourage all studied plant growth characters, chemical composition and therefore yield and its attributes

Table 5: Response of some characters of maize plants to the interaction between irrigation and NPK mineral fertilization treatments

	Normal irrigation		Missing second irrigation		Missing third irrigation		Missing forth irrigation	
Studied characters	NBK 50%	NBK 100%	NBK 50%	NBK 100%	NBK 50%	NBK 100%	NBK 50%	NBK100%
Growth characters								
Plant height/cm	221^{Ba}	259^{Aa}	201^{Bb}	219^{Ab}	188^{Bc}	361.2^{Cc}	127^{Bd}	264^{Cd}
No. of leaves /plant	14^{Ba}	19.3^{Aa}	13^{Bb}	14 ^{Ab}	11.7^{Bc}	81.8^{Cc}	8.7^{Bd}	46.7^{Cd}
Fresh weight g/plant	712^{Ba}	1111^{Aa}	443^{Bb}	687^{Ab}	473.7^{Bc}	480^{Cc}	248^{Bd}	338.6^{Cd}
Dry weight g/plant	127^{Ba}	212^{Aa}	99.1^{Bb}	124.8^{Ab}	79.3^{Bc}	12 ^{Ac}	34.5^{Bd}	10^{Cd}
Leaf area/cm^2	460^{Ba}	616.4^{Aa}	406^{Bb}	453.6^{Ab}	358^{Bc}	188.6^{Bc}	231^{Bd}	141.7^{Cd}
Chemical composition								
Pigment	48.8^{Ba}	55.3 ^{Aa}	44.6^{Bb}	48.6^{Ab}	39.6^{Bc}	41^{Bc}	27.4^{Bd}	33.7^{Cd}
Ch. (μmol m^{-1})	677^{Ba}	837 ^{Aa}	583^{Bb}	672.2^{Ab}	483^{Bc}	510.1^{Cc}	283^{Bd}	378^{Cd}
N (%)	1.57^{Ba}	1.9 ^{Aa}	1.41^{Bb}	1.49^{Ab}	1.19^{Bc}	1.2^{Cc}	0.62^{Bd}	0.88^{Cd}
P (%)	0.85^{Ba}	1.04 ^{Aa}	0.66^{Bb}	0.81^{Ab}	0.58^{Bc}	0.59^{Bc}	0.27^{Bd}	0.5^{Cd}
K (%)	2.5^{Ba}	4.99 ^{Aa}	2.41^{Bb}	2.49 ^{Ab}	2.29^{Bc}	2.31^{Bc}	1.25^{Bd}	1.91^{Cd}
Yield and its components								
Ear weight/g	305^{Ba}	408.3^{Aa}	268^{Bb}	302^{Bb}	215.3^{Bc}	225.7^{Cc}	139^{Bd}	162^{Bd}
No. of grains/ear	478^{Ba}	639.3^{Aa}	418^{Bb}	468.7^{Ab}	238^{Bc}	360^{Cc}	127^{Bd}	215^{Cd}
Bio yield (t/fed)	7.08^{Ba}	9.58^{Aa}	5.98^{Bb}	6.98^{Ab}	4.88^{Bc}	4.98^{Cc}	2.98^{Bd}	3.48^{Bd}
Grain yield (t/fed)	2.98^{Ba}	3.88 ^{Aa}	2.58^{Bb}	2.88^{Ab}	1.98^{Bc}	2.18^{Cc}	1.38^{Bd}	1.58^{Cd}
100 grain weight	22.6^{Ba}	30.8 ^{Aa}	19.6^{Bb}	22.3 ^{Ab}	15.4^{Bc}	16.2^{Cc}	9.3^{Bd}	11.1^{Cd}

Mean having the same capital letters in the same row are not significantly differed at $p \geq 0.05$

Table 6: Response of some growth characters of maize plant to irrigation, bio-fertilization and NPK mineral fertilization treatments

	Normal irrigation			Missing second irrigation			Missing third irrigation			Missing forth irrigation		
Studied characters	0%	50%	100%	0%	50%	100%	0%	50%	100%	0%	50%	100%
Different concentration of NPK mineral fertilization Biofertilizer 1												
Plant height/cm	228.7^{Ea}	253.6^{Ba}	239.7^{Fa}	205.3^{CDb}	218.3^{Ab}	212^{Bb}	195.3^{BCc}	200.6^{Ac}	199^{Ac}	154.7^{Ed}	186^{Ad}	168^{Cd}
No. of leaves/plant	15Da	18.7^{Ba}	15Da	13^{Bb}	14^{Ab}	13^{Bb}	12^{Ac}	12^{Ac}	12^{Ac}	10.7^{Ad}	11^{Ad}	11^{Ad}
Fresh weight g/plant	753^{FFa}	939.3^{Ba}	821Da	577.3^{Bb}	655^{Ab}	602^{Bb}	490^{ABCc}	441^{Ac}	5323^{ABc}	383.7^{BCd}	452.7^{Ad}	433.3^{ABd}
Dry weight g/plant	141.3^{DEa}	168^{Ba}	156.1^{BCc}	106^{Db}	191^{ABb}	110.2^{Cb}	87.7^{ABc}	95.8^{Ac}	91.6^{Ac}	63.5^{BCd}	78.3^{Ad}	71.5^{ABd}
Leaf area/cm^2	479.2^{FFa}	54.72^{Ba}	497.8^{Da}	430.2^{BCb}	443.6^{BCb}	436.3^{BCb}	374.2^{CDc}	402^{ABc}	392^{ABc}	286^{Cd}	353^{Ad}	327^{Bd}
Different concentration of NPK mineral fertilization Biofertilizer 2												
Plant height/cm	224.7^{Ca}	242.3^{Ca}	235Da	204.7^{CDb}	216.3^{Ab}	208^{Cb}	193^{Cc}	199.3^{Ac}	197.3^{ABc}	146.7^{Fd}	176.7^{Bd}	164^{Dd}
No. leaves /plant	14 ^{Ea}	16^{Ca}	15Da	13^{Bb}	13.3^{Bb}	13^{Bb}	12^{Ac}	12 ^{Ac}	12^{Ac}	10^{Bd}	11^{Ad}	11^{Ad}
Fresh weight g/plant	739^{Fa}	879.3^{Ca}	790.7^{DEa}	557.3^{BCb}	655^{Ab}	583^{Bb}	488^{ABCc}	533^{ABc}	517^{ABCc}	369.6^{Cd}	442.7^{Ad}	421.5^{ABd}
Dry weight g/plant	13.2^{Ea}	159.8^{Ba}	146.9^{CDa}	103.2^{Cb}	114.6^{BCb}	107^{CDb}	85.4^{ABc}	92.5^{Ac}	90^{ABc}	55.2^{CDd}	76.6^{Ad}	70^{ABd}
Leaf area/cm^2	466^{FGA}	516.8^{Ca}	485.8^{DEa}	421.3^{Cb}	439.2^{BCb}	435.6^{BCb}	366.6^{CDc}	394^{CDc}	384^{BBc}	276^{CDd}	341^{ABd}	300^{Cd}

	Control concentration of NPK mineral fertilization							
	50%	100%	50%	100%	50%	100%	50%	100%
Plant height/cm	221.3^{Fa}	259^{Aa}	202^{Db}	219^{Ab}	188^{Dc}	188.6^{Dc}	127^{Hd}	141.7^{Gd}
No. leaves/plant	14^{Ea}	19.3^{Aa}	13^{Bb}	14^{Ab}	11.7^{Ac}	12^{Ac}	8.7^{Cd}	10^{Bd}
Fresh weight g/plant	712^{Fa}	1121^{Aa}	542.6^{Cd}	687^{Ab}	473.7^{BCc}	480^{BCc}	248.1^{Dd}	338.6^{Cd}
Dry weight g/plant	127.4^{Ea}	212^{Aa}	109.1^{Db}	124.8^{Ab}	79.3^{Bc}	81.8^{Bc}	34.5^{Ed}	46.7^{Dd}
Leaf area/cm^2	460 ^{Ga}	616.4^{Aa}	406^{Db}	453.6^{ABb}	358^{Dc}	361.2^{Dc}	231^{Dd}	264^{Ad}

Mean having the same capital letters in the same row are not significantly differed at $p \geq 0.05$

compared with either bio-fertilizer-2 or the dosage 50% of NPK. With respect to NPK fertilization treatments under different irrigation treatments Table 5 revealed that under normal irrigation treatment 100% of NPK was the recommended treatment to enhance all studied growth characters, chemical composition, yield and its components when comparing with 50% of NPK.

Concerning with bio-fertilization treatments under different irrigation and NPK fertilization treatments. Data recorded in Table 6, 7 and 8 revealed that the combination of bio-fertilizer-l with 50% of NPK was the superior, since it encouraged significantly all studied characters. Under the moderate water stress, when the second irrigation is absent, the previous combination was capable to overcome all bad effects obtained from water stress and could significantly enhance all studied growth characters, chemical composition, yield and its components. It was noticed that the second recommended treatment under these conditions was

Table 7: Effect of interaction between irrigation, bio-fertilization and NPK mineral fertilization treatments on chemical composition of maize plants

	Normal irrigation			Missing second irrigation			Missing third irrigation			Missing forth irrigation		
Studied characters	0%	50%	100%	0%	50%	100%	0%	50%	100%	0%	50%	100%
Different concentration of NPK mineral fertilization Biofertilizer 1												
Pigment	49.5^{DAa}	54.2^{Aa}	51.1^{Ca}	45.9^{DEb}	48.2^{Abb}	47.2^{Cb}	42^{Dc}	44^{ABc}	42.8^{Cc}	36.2^{CDd}	39.3^{Ad}	38.3^{Abd}
Ch. (μmol m)	688.5^{Ea}	808.2^{Ba}	732.5^{Da}	611^{FGb}	663.8^{BCd}	640.4^{DEb}	528.7^{Dc}	570.4^{ABc}	547^{CDc}	420.4^{CDd}	478^{Ad}	549.1^{Dd}
N (%)	1.64^{Da}	1.89^{Aa}	1.70^{Ac}	1.42^{ABb}	1.47^{ABb}	1.44^{ABb}	1.22^{ABCc}	1.28^{Ac}	1.24^{ABCc}	1.13^{CDd}	1.18^{Abd}	1.16^{Bd}
P (%)	0.89^{CDa}	1.04^{Aa}	0.91^{Ca}	0.68^{Eb}	0.78^{ABb}	0.72^{CDd}	0.60Bc	0.64^{ABc}	0.61^{Bc}	0.54^{Cd}	0.57^{Abd}	0.55^{Bd}
K (%)	2.53^{Ca}	2.79^{Ba}	2.57^{Ca}	2.42^{ABb}	2.46^{ABb}	2.44^{ABb}	2.33^{Ac}	2.40^{Ac}	2.36^{Ac}	2.06^{Abd}	2.24^{Ad}	2.11$^{AB d}$
Different concentration of NPK mineral fertilization Biofertilizer 2												
Pigment	49.1^{EFa}	52.7^{Ba}	50.6^{CDa}	45.1^{DEb}	47.4^{BCb}	46.2^{CDb}	41.7^{Dc}	43.3^{BCc}	42.4^{CDc}	35.3^{Dd}	38.8^{Ad}	37^{BCd}
Ch. (μmol m)	683.5^{EFa}	769.6^{Ca}	730De	594.4^{GHp}	647.2^{CDb}	619.8^{EFb}	524^{Dc}	556.8^{BCc}	538.8^{CDc}	404.1^{Dd}	467.8^{Ad}	434.8^{Cd}
N (%)	1.59^{Fa}	1.8^{Ba}	1.69^{CDa}	1.42^{Bb}	1.45^{ABb}	1.44^{ABb}	1.21^{BCc}	1.26^{ABc}	1.23^{ABCc}	1.11^{Dd}	1.17^{Abd}	1.18^{Abd}
P (%)	0.87^{Da}	0.99^{Ba}	0.90^{Ca}	0.68^{Eb}	0.75^{BCb}	0.70^{DEb}	0.60^{BCc}	0.62^{BCc}	0.61^{Bc}	0.51^{Cd}	0.56^{Bd}	0.55$^{BC d}$
K (%)	2.51^{Ca}	2.69^{BCa}	2.44^{Ca}	2.42^{ABb}	2.44^{ABb}	2.44^{ABb}	2.31^{Ac}	2.40^{Ac}	2.35^{Ac}	1.97^{Bd}	2.16^{Abd}	2.09^{Abd}

	Control concentration of NPK mineral fertilization							
	50%	100%	50%	100%	50%	100%	50%	100%
Pigment	48.4^{Fa}	55.2^{Aa}	46.4^{EFb}	48.6^{ABb}	39.6^{EFc}	41^{DEc}	27.4^{Fd}	33.7^{Ad}
Ch. (μmol m)	676.8^{EFa}	836.6^{Aa}	583.1^{HIb}	672.2^{ABb}	483^{Fc}	510.1^{Ec}	283^{Fd}	377.7^{Ed}
N (%)	1.57^{Ea}	1.90^{Aa}	1.41^{Bb}	1.49^{Ab}	1.19^{Cc}	1.20^{BCc}	0.62^{Fd}	0.88^{Ed}
P (%)	0.85^{Da}	1.04^{Aa}	0.66^{Eb}	0.81^{Ab}	0.58^{BCc}	0.59^{BCc}	0.37^{Ed}	0.50^{Dd}
K (%)	2.5^{Ca}	0.49^{Aa}	2.41^{Bb}	2.49^{ABb}	2.29^{Ac}	2.31^{Ac}	1.25^{Cd}	1.91^{Bd}

Mean having the same capital letters in the same row are not significantly differed at $p \geq 0.05$

Table 8: Effect of interaction between irrigation, bio-fertilization and NPK mineral fertilization treatments on yield and yield components of maize plants

	Normal Irrigation			Missing second irrigation			Missing third irrigation			Missing forth irrigation		
Studied characters	0%	50%	100%	0%	50%	100%	0%	50%	100%	0%	50%	100%
Different concentration of NPK mineral fertilization Biofertilizer 1												
Ear weight/g	324.3^{Ea}	359.3^{Ba}	336.7^{CDa}	277.7^{CDb}	299.7^{Ab}	593.3^{ABb}	243 Cc	260^{Ac}	249.3^{BCc}	137.3^{CDd}	209.7^{Ad}	194.3^{BCd}
No. grains/ear	495.3^{Ea}	562.7^{Ba}	524^{CDa}	439^{DEb}	466^{ABb}	453.3^{CDb}	386.7^{CDc}	417.3^{Ac}	398.7^{BCc}	364^{Dd}	329^{Ad}	289^{BCd}
Bio yield t/fed.	7.41^{Ea}	8.74^{Ba}	8.08^{Da}	6.30^{Eb}	6.87^{Bb}	6.49 Db	5.17 DEc	5.88^{Ac}	5.46 Cc	3.88^{Dd}	4.70^{Ad}	4.39^{Bd}
Grain yield t/fed.	3.45^{Ea}	3.45	3.23^{CDa}	2.66^{CDb}	2.87^{Ab}	2.81^{ABb}	2.33 Cc	2.49^{Ac}	2.39^{BCc}	1.80^{CDd}	2.01^{Ad}	1.86^{BCd}
100 Grain weight	24.11^{Ea}	26.91^{Ba}	25.10^{CDa}	20.37^{CDb}	22.13^{Ab}	21.63^{ABb}	17.6 Cc	18.96^{Ac}	18.11^{BCd}	13.15^{CDd}	14.93^{BCd}	13.71^{BCd}
Different concentration of NPK mineral fertilization Biofertilizer 2												
Ear weight/g	313.3^{Fa}	344^{Ca}	330^{DEa}	273^{DEb}	298.3^{Ab}	285.3^{BCb}	234^{Dc}	253.7^{ABc}	246.3^{BCc}	179^{Dd}	200.7^{Bd}	190.7^{Cd}
No. grains/ear	489.7^{EFa}	537.3^{Ca}	521.7^{Da}	429.7^{Eb}	460^{BCb}	446 DB	377.7^{Dc}	404.7^{ABc}	394^{BCc}	255^{Dd}	303^{Bd}	282^{Cd}
Bio yield t/fed.	7.18^{Ga}	8.40^{Ca}	7.81^{Ea}	6.14 Fb	6.69^{Cb}	6.36^{DEb}	5.06^{FFc}	5.61 Bc	5.29^{Dc}	3.72^{Ed}	4.57^{Ad}	4.07^{Cd}
Grain yield t/fed.	3.00^{Fa}	3.30^{Ca}	3.16^{DEa}	2.62^{DEb}	2.86^{Aa}	2.73^{BCb}	2.24^{Dc}	2.43^{ABc}	2.36^{BCc}	1.72^{Dd}	1.83^{Bd}	1.93^{Cd}
100 Grain weight	23.23^{Fa}	25.68^{Ca}	24.56^{DAa}	20.01^{DEb}	22.03^{Ab}	20.99^{BCb}	16.88^{Dc}	18.45^{ABc}	17.87^{BCc}	12.48^{Dd}	14.21Bd	13.41^{Cd}

	Control concentration of NPK mineral fertilization							
	50%	100%	50%	100%	50%	100%	50%	100%
Ear weight g	305.3^{FGa}	408.3^{Aa}	267.7 Fb	301.7^{Ab}	215.3^{Ec}	225.7^{Dc}	139.3^{Fd}	162^{Fd}
No. grains/ear	478^{FGa}	639.1^{Aa}	418^{EFb}	468.7^{ABb}	338^{Fc}	360^{Ec}	127^{Fd}	215^{Fd}
Bio yield T/fed	7.09^{GHa}	9.56^{Aa}	6.00 Gb	6.96^{ABb}	4.88^{Gc}	4.97^{FGc}	3.07^{Gd}	3.46^{Fd}
Grain yield t/fed	2.93^{FGa}	3.91^{Aa}	2.56^{Eb}	2.89^{Ab}	2.06^{Ec}	2.16^{Dc}	1.33^{Fd}	1.55^{Fd}
100 Grain weight	22.59^{FGa}	30.83^{Aa}	19.57^{Eb}	22.29^{Ab}	15.39^{Fd}	16.21^{Dc}	9.31^{Fd}	11.12^{Fd}

Mean having the same capital letters in the same row are not significantly differed at $p \geq 0.05$

bio-fertilizer-2 with 50% NPK. It was noticed that, when missing the 3rd or the 4th irrigation, plants will face the challenges of the severe water stress conditions. Only bio-fertilizer-l with 50% NPK and bio-fertilizer-2 with 50% NPK were able to overcome the unfavorable growth conditions and could significantly improve all studied growth characters, hence chemical composition, yield and its components.

DISCUSSION

Water is the most abundant constituent of living things. The living tissues of plants usually contain more than 70% by weight of water and maintenance of satisfactory water content is essential for the plant-tissue water content can markedly influence processes of growth and metabolism.

All land plants are to some degree adapted to the unfavorable water regime of their habitat, but some species can tolerate far more unfavorable draught stresses than can other species[45]. Generally there are three basic types of adaptation which can occur, (a) The control of water loss from the plant may be more efficient, (b) The uptake of water may be more efficient and (c) The plant may have a greater ability to grow and metabolize or survive when its tissues are suffering a water deficit[46]. On the other hand[47,48] claimed that water stress influences enzyme activity and in this way can influence all metabolic processes. Moreover, lowering water potential often synthetic processes are reduced more than breakdown processes. He mentioned also, the level of auxin and cytokinins in the tissue are reduced while the level of abscisic acid and ethylene are raised. The auxin change is due to at least partly to increased IAA oxidase activity. Parallely, the period of draught often cause yellowing and later browning of leaves, symptoms similar to senescence. On the other hand, the tolerance to draught under field conditions was studied in barely varieties. It was found that higher proline accumulation during draught were the more tolerant to draught. Muhammad[49], Derby *et al.*[4] and Barnabas *et al.*[50] claimed that the unfavorable growth conditions such as water stress, salinity or even heat stress can be tolerated by plants in juvenility rather than those at maturity. This is because plants in juvenile have high concentration of growth promoters such as IAA, GA and CKs. It helps significantly in compensating any decrease happen in photosynthesis pathway, mineral absorption and production of inhibitors such as ethylene and ABA when stressed occurred. On the other hand, Devlin[51] and Al-Kaisi and Yin[52] reported that plants at maturity generally have high concentrations of the inhibitors comparing with the promoters this encourages assimilates transportation from sources to sinks accompanied with fruity parts. The previous discussion clarify the results obtained in this study, taking into consideration the hazard effects of water stress on maize plants growth, chemical composition and hence yield and its components especially at the end of the juvenility compare with the early juvenile growth period. Moursi *et al.*[53], Ibrahim *et al.*[54], Moursi *et al.*[55], Reaid *et al.*[8], Chen *et al.*[56], Li *et al.*[57] and Schluter *et al.*[58] reported that maize plant is considered as one of the greediest crops to use fertilizers particularly to nitrogen. Nitrogen considered as one of the major elements which enhance assimilates metabolism and transportation by enhancing plant photosynthesis rate. This can explain the obtained observations as a result of adding the full dosage of NPK fertilization when compared with the half dosage fertilization or with applying both of the bio-fertilizers under study. It could be stated that the biological fertilization may considered as the only available solution to apply as a fertilizers when there is an irrigation water scarcity. Vargas *et al.*[59] and Nur *et al.*[60] stated that positive plant growth responses after inoculation with associated N_2 fixing bacteria were found under water stress conditions. The highest results obtained when applying bio-fertilizer-l rather than bio-fertilizer-2 may be achieved as a consequence of the microorganisms efficiency in N-fixation, production of organic acids and phytohormones as IAA, GA3 and CKs, which led to increase P and K availability in plants rhizosphere, in addition to the higher P and K release capability of the micro-organisms in bio-fertilizer-l if compared with bio-fertilizer-2. These results are in concomitant with Ishac[61] and Schroeder and Janos[22]. We can state that all mentioned factors together led to produce higher yield as a result of incurring plant growth and this led to improve plant chemical composition and metabolism. The superior results obtained from bio-frtilizer-1 either under normal irrigation or water stress conditions, were because of the hyphal development of the Mycorrhiza which play a big role in improving the soil mechanical texture out of the nature of fungal growth; also it plays as lateral roots exchanging the carbohydrate and the amino acids from the co-operated plant to the fungi, on the other hand, phosphate and other minerals from the fungus to the co-operated plant. Under water stress conditions, the hyphal of Mycorrhiza play as an additional lateral root system providing the water from long distances away from the root system to the plants[62,63]. Moreover, Jakobsen *et al.*[64] and Harley[65] claimed that the increase in nutrient uptake may be due to the physical increase in the surface area for nutrient uptake. This is partly due to the mycelium in the soil having a much greater surface area and extends more than root hairs. In addition, the fungus infection prolongs the life of lateral rootlets. *Azotobacter crococcu*, as the second micro-organism in the biofertilizer-1, helped in increasing nitrogen soil content through non-symbiotic nitrogen fixation pathway. This led to the production of plant growth promoting phyto-hormones such as IAA, GA3 and CKs, which helped in encouraging plant growth and organic acids therefore reducing soil pH, thus release the unavailable soil nutrients particularly zinc and phosphate especially under calcareous soil conditions. All these factors together led to enhance the photosynthetic pigments accumulation thus increase photosynthesis pathway as well as increased yield and its components[21,51,61,66]. When NPK soil content is increased through mineral fertilization, the micro-organisms in both bio-fertilizers used the suitable NPK which added directly into soil instead of going through N-fixation. Unavailable P and K release pathway, by the mean of micro-organisms inhibition because of high soil content of NPK.[22,61,67]. This can illustrate the results observed under all studied water regime conditions. But once discussing the severe water stress conditions when missing 3rd or the 4th irrigation, plants faced complex challenges which seriously defend against implementing plant life cycle. Bio-fertilization remains alone capable to overcome the water stress hazard effects through producing growth promoting phyto-hormones such as IAA, GA and CKS, beside the organic acids. Moreover, activate the enzyme phosphatase which helps in releasing P and K in plant rhizosphere. Consequently, encourage the plant metabolism and improve the plant chemical compositions and growth, to be confirmed finally as significant increase in yield and its components[68,69].

CONCLUSION

The biological fertilization technique considered as biofertilizer and a source of NPK is urgently needed to save the environment and reduce the running costs of crops production particularly in the new reclaimed areas and to avoid the inapplicable risks of applying the mineral fertilization under water stress conditions. Using the biofertilizer-I alone is highly recommended under water stress conditions, while it is permissible accompanied with the half of the recommended dosage of mineral NPK when irrigation water is sufficient enough.

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