

## The Beneficial Effect of Biofertilizer Together with Ascorbic Acid on Roselle Plants Grown Below Different Kinds of Soil

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**Abstract:** This study was conducted to evaluate the impact of bio fertilizers namely, Azotobacterine (*Azotobacter chroococcum*) and phosphorein (*Bacillus polymyxa*) and mineral N, P and K fertilizers at the rates of 25, 50 and 100% for each fertilizer (from the doses recommended by Ministry of Agriculture) as well as ascorbic acid at the rate of 400 ppm on growth (plant height, number of branches and herb fresh and dry weights/plant), yield (dry weight of sepals/plant and feddan and weight of seeds/plant and feddan) of roselle plants (*Hibiscus sabdariffa*, L.) under different soils (clay soil at Dar El-Ramad farm, sandy loam and saline loamy sand soil at Demo farm, Faculty of Agriculture) at El-Fayoum governorate conditions. The data obtained showed that, bio and mineral (NPK) fertilizers increased the above characters of roselle plants under different soils of experiment. The maximum increase of these characters was obtained by the treatment clay soil × 100% NPK + biofertilizers × 400 ppm ascorbic acid, followed by clay soil × 50% NPK + biofertilizers × 400 ppm ascorbic acid as compared to saline loamy sand soil × non fertilizer × zero ascorbic acid treatment, although, the differences between these treatments and mineral fertilizer at the rate of 100% NPK alone were insignificantly. Therefore, it is economically and environmentally recommended to inoculate roselle seeds with mixture of Azotobacter + Bacillus and fertilize these inoculated plants with 50% plus 400 ppm ascorbic acid improve the vegetative growth and augment the yield components of roselle plants under clay soil with spraying 400 ppm of ascorbic acid were obtained the best results of this work study.

**Keywords:** Nitrogen, phosphorus, Azotobacterine, phosphorein, antioxidant, salinity.

### INTRODUCTION

Roselle (*Hibiscus sabdariffa*, L.) is cultivated mainly in Upper Egypt and grows well under different environmental conditions such as high temperature [1]. Roselle requires a permeable soil, a friable sandy loam with humus being preferable. The cultivated area of roselle plants at El-Fayoum governorate reached to more than 100 feddans. The new reclaimed soils can be cultivated with such plants, which are able to grow under different climatic and soil type conditions [2]. Plants need certain amount of bound nutrients in specific type at acceptable time, for her growth and development [3]. Recently, great efforts have been done on the use of biofertilizers instead of chemical ones to produce clean and healthy crops. The use of mineral fertilizer is the major cost in plant production and causes the environmental pollution, as well as, contaminates the underground water [4]. Biofertilizers mainly consist of beneficial microorganisms that can release nutrients and make them available in the soil for growing economical plants. Besides, bio-fertilization has the advantages of avoiding environmental pollution and being cheap. Soil microorganisms which convert the insoluble form of phosphorus to soluble for play an

important role in supplying the plants with the available phosphorus [5]. Phosphorein is a biofertilizer product containing active microorganisms hydrolyzing the insoluble phosphorus into soluble one. Also, phosphate solubilizing bacteria solubilize insoluble P by producing various organic acids. This available P is taken up by plants [6&7]. In addition, Azotobacterine, Microbiene, Nitrobiene, and Biogen as new bio-fertilizers have greater amount of symbiotic and no symbiotic bacteria responsible for fixation of nitrogen by atmosphere. Application of them achieved the following merits (According to [8,9&10]); reducing plant requirement of N by 25%, and increasing the availability of various nutrients, stimulating growth of roots, enhancing the resistance of plants to root diseases, reducing the environmental pollution, and improving the productive performance of the fruits crops.

Ascorbic acid is an important antioxidant defense in plant cells [11]. It protects plant cells against damage by oxygen free radicals, which may be produced as a result of a disturbance of electron transfer processes or via autooxidation. It also stimulated respiration activities, cell division and many enzymes

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activities. Recently, a great attention has been focused on the possibility of using natural and safety substances i.e. vitamins, amino acids and yeasts in order to improve plant growth, flowering, capsule setting and resistance against unfavorable environmental conditions and pathogens [12&13]. However, lack of information about the physiological roles of such factors is still exists. Foliar application of ascorbic acid to different plant species was found to have stimulative effects on the growth, dry matter accumulation and yield components.

A general reduction in growth and yield due to salinity is widely documented [14]. Growth and yield of roselle plants were decreased with increasing soil salinity [15]. The drastic influence of salinity on plant growth and metabolism was attributed, principally, to the enhanced  $\text{Na}^+$  uptake which causes ion excess in plant tissues [16]. One of the primary effects of increasing salinity in the growth medium is the inhibition of  $\text{K}^+$ ,  $\text{Ca}^{++}$  and  $\text{NO}_3^-$  uptake by plant roots [17]. In addition it is well established that salinity stress damages plant cells through production of reactive oxygen species including superoxide, hydrogen peroxide, hydroxyl anins and single oxygen [18]. On the other hand, some trials have been made to alleviate the disturbances in plant metabolism excreted by salinity stress. It has been suggested that some

antioxidants (to which belongs ascorbic acid) may help to overcome some of these inhibitory effects.

The objective of this work is to investigate the response of roselle plants (*Hibiscus sabdariffa*, L.) to different levels of mineral, bio fertilization, ascorbic acid as well as, their interactions on plant growth, yield and chemical composition in order to figure out the possibility of eliminating or/and decreasing the amount of major chemical fertilizers by substitution with biofertilizers and ascorbic acid under El-Fayoum governorate conditions.

## MATERIAL AND METHODS

A field experiment were conducted throughout the 2 sequential seasons of "2007" and "2008" to check the result of bio, nitrogen, phosphorus and potassium fertilizers on growth, yields and its part of roselle plants (*Hibiscus sabdariffa*, L.) underneath completely different soils at El-Fayoum governorate conditions. Three completely different soils in two experimental stations of the Faculty of Agriculture, particularly clay soil at the Dar El-Ramad farm, a saline loamy sand and sandy loam soil at Demo farm. Some chemical and physical characteristics of the experimental stations soils throughout "2007" and "2008" seasons are given in Tables (1).

**Table-1: Some physical and chemical properties of Dar El-Ramad and Demo farms through season 2007 and 2008**

Soil properties	Season 2007			Season 2008		
	Demo		Dar El-Ramad	Demo		Dar El-Ramad
<b>Physical properties</b>	28.69	30.20	5.14	22.73		6.37
Coarse sand %	37.19	42.33	21.55	31.66	31.05	19.40
Fine sand %	22.16	15.72	24.43	31.84	40.38	27.48
Silt %	11.96	11.75	48.88	13.77	16.04	46.75
Clay %	<b>Sandy loam</b>	<b>Loamy sand</b>	<b>Clay</b>	<b>Sandy loam</b>	<b>Loamy sand</b>	<b>Clay</b>
Texture class						
<b>Chemical properties</b>	0.48	0.41	1.69	0.38	0.40	1.43
Organic matter %	8.61	7.42	5.82	7.83	7.69	5.12
$\text{CaCO}_3$ %	7.82	7.76	7.89	7.75	7.81	7.55
pH (soil paste)	3.12	7.51	1.58	3.64	7.26	2.10
E <sub>Ce</sub> (paste extract), ds/m						
Soluble anions, meq/L*	....	....	....	....	....	....
$\text{CO}_3^{--}$	5.40	4.37	2.24	4.83	4.96	3.83
$\text{HCO}_3^-$	12.35	38.93	7.12	15.42	35.43	10.29
$\text{Cl}^-$	13.45	31.80	6.44	16.15	32.21	6.88
$\text{SO}_4^{--}$						
Soluble cations, meq/L*	8.67	30.75	6.15	10.25	30.24	7.43
$\text{Ca}^{++}$	6.89	17.41	6.80	7.32	14.39	8.92
$\text{Mg}^{++}$	15.03	26.15	2.13	18.11	27.32	3.80
$\text{Na}^+$	0.61	0.79	0.72	0.72	0.65	0.85
$\text{K}^+$						
Available nutrients, ppm**						
N	3.54	4.44	21.02	3.80	4.96	22.16
P	6.87	6.42	12.27	6.59	6.42	12.27
K	54	36	83	42	24	89

\*In saturation paste extract, \*\*On dry weight basis

Biofertilizers specifically Azotobacterine (*Azotobacter chroococcum*) as nitrogen fixing bacterium and phosphorein (*Bacillus polymyxa*) as a phosphate dissolving bacterium were obtained from Agricultural

Center, Ministry of Agriculture, Egypt. The seed of roselle cv. Sabahia 17 were obtained from the Research Center of Medicative and Aromatic Plants, Ministry of Agriculture, Egypt. Seeds were immersed in Arabic gum solution (16%) as a protrusive agent, then, the seeds were mixed with the powder of mixed biofertilizers; inoculated seeds were allowed to dry

before sowing. Application of N, P and K fertilizers were a hundred, fifty and twenty fifth for every fertilizer from that counseled doses by the Ministry of Agriculture, five hundred weight unit ammonia salt (20.6% N), one hundred fifty weight unit calcium superphosphate (15% P<sub>2</sub>O<sub>5</sub>) and fifty weight unit potassium sulfate/fed (48% K<sub>2</sub>O). Nitrogen fertilizer was applied in 2 equal doses, the primary dose throughout sowing and before the primary irrigation, and therefore the second dose once one month from sowing. Phosphorus and potassium fertilizers were applied throughout soil preparation.

Application of mineral fertilization	N	P	K
	Ammonium sulfate (20.6% N)	Calcium superphosphate (15% P <sub>2</sub> O <sub>5</sub> )	Potassium sulfate (48% K <sub>2</sub> O)
100%	500 kg/fed	150 kg/fed	50 kg/fed
50%	250 kg/fed	75 kg/fed	25 kg/fed
25%	125 kg/fed	37.5 kg/fed	12.5 kg/fed

### Ascorbic acid

Ascorbic acid at rates of zero and 400 ppm were sprayed on roselle plants at the two times, 60 and 90 days from the sowing. Some drops of liquid soap were added to ascorbic acid solution as wetting agent. Plants were sprayed in the early morning with the ascorbic acid solution (300 liter per feddan) from the leaves. The untreated plants were sprayed with the same volume of tap water.

### Treatments

The experiment included thirty six treatments with three replicates (three different actual soils × two levels of ascorbic acid × six levels of NPK fertilizers).

### The treatments were as following

- 1- Non fertilizer × Zero ascorbic acid.
- 2- Non fertilizer × 400 ppm ascorbic acid.
- 3- Biofertilizers × Zero ascorbic acid.
- 4- Biofertilizers × 400 ppm ascorbic acid.
- 5- 100% NPK × Zero ascorbic acid.
- 6- 100% NPK × 400 ppm ascorbic acid.
- 7- (Biofertilizers + 100% NPK) × Zero ascorbic acid.
- 8- (Biofertilizers + 100% NPK) × 400 ppm ascorbic acid.
- 9- (Biofertilizers + 50% NPK) × Zero ascorbic acid.
- 10- (Biofertilizers + 50% NPK) × 400 ppm ascorbic acid.
- 11- (Biofertilizers + 25% NPK) × Zero ascorbic acid.
- 12- (Biofertilizers + 25% NPK) × 400 ppm ascorbic acid.

These treatments were repeated with each one of soil type. Soils were in the main plots, ascorbic acid treatments in the sub-plots and fertilization treatments in the sub-sub-plots. This design offers a good opportunity for obtaining an accurate idea about the optimum level for any main factor as each was applied

in the different treatments. Also, the first and second order interactions could be determined.

The experimental field was plowed, rolled and divided into fifty four plots each one of 10.5m<sup>2</sup> area (1/400 fed) and contains five rows (3 m dimension and 3.5 m in length). The seeds inoculated were seeded on frist and second May, within the 2 experimental seasons. Four seeds were seeded per hill (50 cm apart) on one facet of the ridge. Once one month from sowing the seedlings were diluted to one plant per hill. During this regard, every experimental unit (plot) contains thirty plants (6plants/row) and 12000 plants/fed. All alternative scientific discipline practices were followed as suggestion in roselle management.

### Plant samples

Samples of roselle plants (9 plants) were every which way chosen from every treatment (3 plants for every replicate) within the early morning when a hundred and forty days from sowing now transferred to the laboratory to check the subsequent morphological characteristics and plant analysis.

1. Average plant height (cm) was measured from the cotyledonary node to plant high of the most stem.
2. Average range of branches/plant: was calculable by taking the typical range of branches of 9 plants.
3. Average recent weight (g): The samples were stemmed by taking the typical weight of 9 plants.
4. Average dry weight of herb g/plant: The samples were dried in an electrical oven appliance at seventy °C ± two till constant weight was reached nearly when forty eight hours

### Yield and its components

At the age of a hundred and eighty days from sowing, hibiscus plants were harvested (9 plants) from

every treatment that arbitrarily chosen to check the subsequent parameters.

1. Dry weight of sepal's g/plant: The dry weight of sepals plant (calyx and epicalyx) was recorded when drying in sunny place.
2. Dry sepals yield (kg/feddan): The capsules were air-dried in sunny place. The dry yield of sepals was calculated (kg/fed).
3. Weight of seeds/plant (g): The capsules were collected and dried on paper sheet in sunny open place. When drying, the seeds were separated and its dry weight was recorded.
4. Weight of seeds (kg/feddan): The weight of seeds/feddan was calculated from the burden of seeds/plot.

## STATISTICAL ANALYSIS

Results were statistically analyzed victimization the L.S.D. at chance level of 5% for comparisons according to [19].

## RESULTS

### A- Growth characters

Data presented in Table (2) clearly indicate that the clay soil gave the highest growth parameters of roselle plant, (i.e. plant height, number of branches, herb fresh weight Kg/plant and herb dry weight g/plant) compared to sandy loam and saline loamy sand soil. The application of mineral fertilization (100% NPK) on roselle growth parameters was significantly increased compared to the non-fertilizer in the first and second seasons. Data also show that bio fertilization treatment significantly increased roselle growth parameters in the

first and second seasons, in comparison to non-fertilizer treatment respectively. Seeds inoculation with bio-N&P fertilizer in combination with 25, 50 and 100 % from recommended N&P dose as soil application improved the studied growth parameters of roselle plant, as compared to non-fertilizer treatment. Such trend was true during the two studied seasons. The interaction between mineral fertilizers (NPK) and bio fertilizers significantly affected the roselle growth parameters in the two experimental seasons. All growth parameters for plants that received 100% NPK and inoculated with different bacteria strains were increased than those which did not received neither mineral NPK nor bio fertilization treatments. The most effective interaction treatments were 100% NPK + bio fertilizers followed by 50% NPK + bio fertilizers than 25% NPK + bio fertilizers. The interaction of growth parameters between soil type and fertilizer treatments was increased at the treatment (clay soil × Bio + 100% NPK) in the first and second seasons, respectively above the lowest values at the treatment (saline loamy sand soil × non fertilizer).

The interactions effects between all treatments (soil type × ascorbic acid × fertilization) were significantly affected the growth parameters of roselle plants in the experiments of two seasons. The maximum increases of growth parameters were at the treatment (clay soil × 400 ppm ascorbic acid × Bio + 100% NPK) over the treatment (saline loamy sand soil × zero ascorbic acid × non fertilizer) in the first and second seasons, respectively.

**Table-2: Effect of bio and mineral fertilization as well as ascorbic acid and their interactions on growth parameters (plant height, number of branches, herb fresh weight Kg/plant and herb dry weight g/plant) of roselle plants under clay, sandy loam and saline loamy sand soils during the two successive seasons 2007 and 2008**

Treatments	Plant height (cm)											
	2007						2008					
	Clay		Sandy loam		Saline loam sand		Clay		Sandy loam		Saline loam sand	
	Ascorbic acid						Ascorbic acid					
	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm
Non fertilizer	106.3	123.7	97.0	108.3	77.3	93.7	113.3	125.0	94.0	105.3	77.3	93.7
Biofertilizers	148.0	156.3	131.0	143.0	118.3	135.0	147.7	158.0	130.7	140.0	118.3	135.0
100% NPK	183.7	191.3	174.7	182.0	128.7	150.3	186.3	190.0	172.0	180.0	128.7	150.3
Bio + 25% NPK	174.0	180.3	152.0	162.7	144.3	155.7	175.3	185.7	152.0	164.7	144.3	155.7
Bio + 50% NPK	190.7	193.0	175.0	183.0	153.7	176.3	192.7	197.7	176.7	182.0	155.3	186.3
Bio + 100% NPK	193.0	195.7	177.0	181.0	161.0	178.0	195.3	203.0	172.7	184.0	166.7	182.0
Mean	165.9	173.4	151.1	160.0	130.6	148.2	168.4	176.6	149.7	159.3	131.8	150.5
L.S.D. at 5%	S=27.70 F=8.1 A= 10 S.F.A=3.3						S=7.2 F=6.1 A=11.6 S.F.A=2.4					
Treatments	Number of branches											
	2007						2008					
	Clay		Sandy loam		Saline loam sand		Clay		Sandy loam		Saline loam sand	
	Ascorbic acid						Ascorbic acid					
	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm
Non fertilizer	10.7	12.3	8.3	10.3	9.3	10.3	11.3	11.3	8.0	10.0	9.0	11.0

<b>Biofertilizers</b>	11.7	14.3	11.0	13.7	12.3	14.0	12.3	14.0	11.7	14.0	12.8	15.0
<b>100% NPK</b>	19.7	21.0	18.3	20.0	19.2	18.7	19.3	20.3	18.7	20.3	19.5	19.0
<b>Bio + 25% NPK</b>	15.0	17.7	12.0	15.3	13.7	14.0	14.7	18.0	12.3	15.7	14.0	14.7
<b>Bio + 50% NPK</b>	19.0	21.7	17.3	18.7	18.0	18.7	18.7	21.0	17.7	19.0	18.3	19.0
<b>Bio + 100% NPK</b>	20.3	21.3	16.0	17.0	16.5	17.0	20.7	21.3	16.0	19.0	17.5	17.3
<b>Mean</b>	16.1	18.1	13.8	15.8	14.8	15.4	16.2	17.7	14.1	16.3	15.2	16.0
<b>L.S.D. at 5%</b>	S=3.2 F=3.2 A=2.4 S.F.A=N.S.						S=3.0 F=1.7 A= 2.8 S.F.A=0.7					
<b>Treatments</b>	<b>Herb fresh weight Kg/plant</b>											
	2007						2008					
	Clay		Sandy loam		Saline loam sand		Clay		Sandy loam		Saline loam sand	
	Ascorbic acid						Ascorbic acid					
	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>
<b>Non fertilizer</b>	1.40	1.54	1.29	1.36	1.03	1.29	1.37	1.50	1.16	1.33	0.99	1.24
<b>Biofertilizers</b>	1.85	1.96	1.56	1.58	1.37	1.62	1.79	1.93	1.48	1.50	1.27	1.59
<b>100% NPK</b>	2.13	2.36	1.95	2.12	1.71	1.99	2.09	2.36	1.89	1.98	1.66	1.94
<b>Bio + 25% NPK</b>	1.87	1.93	1.72	1.82	1.47	1.57	1.82	1.88	1.71	1.79	1.40	1.53
<b>Bio + 50% NPK</b>	2.17	2.41	1.89	1.99	1.69	1.87	2.12	2.37	1.84	1.94	1.64	1.84
<b>Bio + 100% NPK</b>	2.22	2.42	1.99	2.01	1.80	1.95	2.17	2.33	1.94	2.00	1.76	1.89
<b>Mean</b>	1.94	2.10	1.73	1.82	1.51	1.72	1.89	2.06	1.67	1.75	1.45	1.67
<b>L.S.D. at 5%</b>	S=2.14 F=0.98 A= 1.51 S.F.A= 4						S=1.22 F=1.2 A= 1.56 S.F.A=4.6					
<b>Treatments</b>	<b>Herb dry weight g/plant</b>											
	2007						2008					
	Clay		Sandy loam		Saline loam sand		Clay		Sandy loam		Saline loam sand	
	Ascorbic acid						Ascorbic acid					
	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>	<b>Zero</b>	<b>400 ppm</b>
<b>Non fertilizer</b>	312	344	293	317	236	294	305	334	259	296	221	277
<b>Biofertilizers</b>	412	437	348	361	313	370	398	429	329	335	284	355
<b>100% NPK</b>	475	525	435	483	389	453	466	525	421	440	370	432
<b>Bio + 25% NPK</b>	416	429	383	415	335	358	405	418	381	398	312	341
<b>Bio + 50% NPK</b>	482	537	422	453	385	426	471	527	409	431	365	409
<b>Bio + 100% NPK</b>	493	538	453	458	411	445	483	519	431	445	393	421
<b>Mean</b>	432	468	389	414	345	391	421	459	372	391	324	372
<b>L.S.D. at 5%</b>	S=45 F=22 A=36 S.F.A=9						S=25 F=23 A=35 S.F.A=9					

## B- Yield and its components

Data presented in Table (3) showed that clay soil gave the best yields and its components (Sepals dry weight g/plant, Sepals dry weight kg/fed, Weight of seeds g/plant and Weight of seeds kg/fed) as compared to sandy loam and saline loamy sand soil in the first season and second season, respectively. The maximum results has been recorded on yields and its components by 100% mineral NPK treatment compared to unfertilized plants as listed in Table (3). In regard to bio fertilization, data also show that application of bio fertilization (with two bacteria species) led to significant increase in yields and its components in comparison with non-fertilizer in the experiments of two seasons. The combination between NPK and bio fertilizers significantly affected yields and its components in the experiments of the two seasons. The most effective interaction treatments were 100% NPK + bio fertilization followed by 50% NPK + bio fertilization. Data tabulated in Table (3) show that the

interaction between soil type and fertilizer treatments greatly increased yields and its components at the treatment (clay soil × Bio + 100% NPK) above the lowest values, at the treatment (saline loamy sand soil × non fertilizer) in the first and second seasons, respectively.

The interaction effect between fertilizer treatments and ascorbic acid (F×A) significantly affected yields and its components in the experiments of the two seasons. The most effective interaction treatments were 100% NPK+ ascorbic acid + bio fertilization followed by 50% NPK + ascorbic acid + bio fertilization than 25% NPK + bio fertilization. The maximum value was obtained by the treatment (100% NPK × 400 ppm ascorbic acid x bio fertilization) while, the lowest value was obtained by the treatment (non fertilizer × zero ascorbic acid) in the first and second seasons.



**Table-3: Effect of bio and mineral fertilization as well as ascorbic acid and their interactions on yields and its components (sepals dry weight g/plant, sepals dry weight kg/fed, weight of seeds g/plant and weight of seeds kg/fed) of roselle plants under clay, sandy loam and saline loamy sand soils during the two successive seasons 2007 and 2008.**

Treatments	Sepals dry weight g/plant																							
	2007						2008																	
	Clay		Sandy loam		Clay		Sandy loam		Clay		Sandy loam													
	Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid													
	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm												
Non fertilizer	21.7	25.3	17.4	19.4	12.9	19.6	18.0	23.3	15.5	19.1	11.6	19.1												
Biofertilizers	32.2	32.8	19.3	21.9	14.0	23.4	28.6	31.2	17.3	21.2	12.8	21.2												
100% NPK	47.7	53.2	23.5	25.3	20.5	27.5	43.6	50.2	21.9	25.4	19.2	25.4												
Bio + 25% NPK	36.5	38.3	21.5	22.9	18.5	24.1	34.1	37.3	18.6	22.4	16.7	22.4												
Bio + 50% NPK	45.3	52.7	23.2	24.6	20.4	24.6	40.6	50.1	20.8	24.6	18.5	24.6												
Bio + 100% NPK	49.4	54.7	26.2	28.0	23.1	24.6	45.1	53.8	24.2	27.1	20.6	27.1												
Mean	38.8	42.8	21.8	23.7	18.2	24.0	35.0	41.0	19.7	23.3	16.6	23.3												
L.S.D. at 5%	S=3.4			F=2.6			A=3.2			S.F.A=0.9			S=2.1			F=2.4			A=3.2			S.F.A=0.6		
Treatments	Sepals dry weight kg/fed																							
	2007						2008																	
	Clay		Sandy loam		Saline loam sand		Clay		Sandy loam		Saline loam sand													
	Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid													
	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm												
Non fertilizer	261	303	209	232	154	236	216	280	185	229	139	236												
Biofertilizers	386	394	232	262	168	281	343	375	208	255	154	270												
100% NPK	573	638	281	304	246	330	523	603	263	305	230	329												
Bio + 25% NPK	438	459	258	274	222	289	410	447	223	269	200	288												
Bio + 50% NPK	543	633	278	295	245	295	487	601	250	295	222	294												
Bio + 100% NPK	592	656	314	336	278	295	542	646	290	325	248	276												
Mean	465	514	262	284	219	288	420	492	237	280	199	282												
L.S.D. at 5%	S=38			F=37			A=41			S.F.A=11			S=16			F=32			A=39			S.F.A=7		
Treatments	Weight of seeds g/plant																							
	2007						2008																	
	Clay		Sandy loam		Saline loam sand		Clay		Sandy loam		Saline loam sand													
	Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid													
	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm												
Non fertilizer	39.9	40.0	18.8	20.8	14.8	20.2	30.3	20.2	13.6	17.5	12.6	17.5												
Biofertilizers	42.4	47.2	21.8	26.6	21.0	25.6	36.4	25.6	18.0	23.3	17.5	24.4												
100% NPK	52.2	53.1	30.5	35.5	28.9	33.7	47.1	33.7	26.3	35.1	25.3	32.2												
Bio + 25% NPK	49.0	52.4	28.5	30.0	26.4	29.8	41.2	29.8	23.9	29.5	22.7	27.4												
Bio + 50% NPK	53.7	54.8	28.0	32.8	27.3	31.0	47.8	31.0	23.4	34.2	23.6	28.5												
Bio + 100% NPK	54.1	55.9	35.8	45.6	32.1	34.4	49.5	34.4	29.2	42.4	28.7	37.7												
Mean	48.5	50.6	27.3	31.9	25.1	29.1	42.1	29.1	22.4	30.3	21.7	27.5												
L.S.D. at 5%	S=16.9			F=4.9			A=3.1			S.F.A=2.0			S=4.9			F=2.8			A=4.9			S.F.A=1.2		
Treatments	Weight of seeds kg/fed																							
	2007						2008																	
	Clay		Sandy loam		Saline loam sand		Clay		Sandy loam		Saline loam sand													
	Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid		Ascorbic acid													
	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm	Zero	400 ppm												
Non fertilizer	478	480	225	250	177	243	363	598	163	373	151	349												
Biofertilizers	509	567	262	319	252	307	437	429	216	210	210	210												
100% NPK	613	637	367	427	347	404	566	502	315	279	304	293												
Bio + 25% NPK	577	589	342	360	317	358	494	585	286	421	272	387												
Bio + 50% NPK	604	642	337	354	328	372	574	526	281	353	283	329												
Bio + 100% NPK	653	671	430	527	385	413	594	592	351	410	344	343												
Mean	572	598	327	373	301	349	505	613	269	508	261	417												
L.S.D. at 5%	S=140			F=49			A=38			S.F.A=20			S=58			F=34			A=59			S.F.A=14		

## DISCUSSION

Uses of bio-fertilizers containing useful microorganisms rather than artificial chemicals square measure far-famed to boost plant growth through the provider of plant nutrients and will facilitate to sustain environmental health and soil productivity. The useful result of immunization of hibiscus seeds with bio-fertilizers N & P chemical combined with twenty five, fifty and 100 percent of the counseled N&P dose from identical nutrient as soil application, resulted in vigorous growth additionally as extremely productivity of plants with smart quality. Moreover, the importance or the prevalence for the applications of bio chemical wasn't solely taken as a criterion for increasing the financial gain for hibiscus crop or rationalize of expensive mineral-N&P fertilizers, however conjointly for minimizing the probably adverse fears of each human health and environmental risks resulted from uncontrolled use of mineral-N fertilizers [20]. An increase in every of growth characters (plant height, etc...) was because of several factors like (a) its ability to unharness plant promoting substances (mainly IAA, gibberellin and plant hormone like substances) which could be stirred up plant growth [21], (b) synthesis of some vitamins, e.g. B12 [22], (c) increasing the water and mineral uptake from the soil [23]. This could be ascribed to extend in root area, root hairs and root elongation as littered with *Azotobacter* [24], (d) increasing the power to convert  $N_2$  to  $NH_4$  and therefore create it offered to plant [25] and (e) enhancing the assembly of biologically active fungistatical substances which can amend the microflora within the rhizosphere and have an effect on the balance between harmful and useful organisms [26].

Some microorganism referred to as Plant Growth Promoting Rhizobacteria (PGPR), stimulate plant growth [27]. The stimulatory effects of microorganisms might result from either direct or indirect action. Direct effects embody production of phytohormones [28], sweetening of convenience of some minerals [27], liberation of phosphates and micronutrients, independent organic process and stimulation of disease-resistance mechanisms [29]. Indirect effects arise from (PGPR) fixing the basis setting and ecology [30]. The rise in measured growth characters (plant height etc...) was thanks to that this treatments leading to a lot of availableness of chemical element to be absorbed by the plants. The positive impact of mineral-N fertilization on growth characters of plants is also attributed to the encourage role of chemical element in substance formation, proteins, amino acids, nucleic acids, several enzymes and energy transfer materials [31]. Nitrogen conjointly fast each organic process and elongation thanks to its nice action in stimulating nutritional standing and therefore the growth parameters. The up impact of N as foliar application on yield and its nitrogen was in the main attributed to its positive action on enhancing

growth parameters (Table 6, 7, 8&9). This successively will increase the quantity of metabolites synthesized by plants [32]. Nitrogen conjointly fast each cellular division and elongation thanks to its nice action in stimulating organic process standing and also the growth parameters. The rising impact of applying N as soil application on yield and its parts was chiefly attributed to its positive action on enhancing growth parameter (Table 6, 7, 8&9). This successively will increase the quantity of metabolites synthesized by plants [32]. Furthermore the positive impact of phosphate dissolving microorganism on growth may well be attributed to the impact of those microorganism to encourage plant to provide some growth promoting substances like auxine, gibberellins and cytokinens [33] which can improve plant growth and stimulate microorganism development within the rhizosphere [34], and their positive action on mineralization and solubilization potential for organic and inorganic phosphorus, respectively [35]. Moreover, it absolutely was found that the applying of phosphate dissolving microorganism as a biofertilizer resulted in a very reduction of soil pH scale that augmented the solubility of some nutrients like P, Fe, Zn, Mn and copper that successively augmented nutrient uptake by plants [36]. Data in Table (2,3,4,5,6,7,8 and 9) show that treated plants by antioxidant (400 ppm) considerably accumulated growth and yield similarly as improved quality of roselle plant compared to untreated plants within the initial and second seasons, severally. The useful result of antioxidant (ascorbic acid) on growth and yield similarly as improved quality of many plants was rumored by many employees such as [37-43] on completely different decorative plants together with roselle plants. Finally, from the obtained data, it could be concluded that cultivation of inoculated seeds of roselle plants by *Azotobacter* and *Bacillus* greatly improved the vegetative growth, yield and some chemical compositions, particularly, in combination with mineral fertilizer and spray with ascorbic acid under different soil types. The maximum increase was obtained by the treatment of clay soil  $\times$  100% NPK + biofertilizer  $\times$  400 ppm ascorbic acid followed by the treatment clay soil  $\times$  50% NPK + biofertilizer  $\times$  400 ppm ascorbic acid, although, the differences between these treatments and mineral fertilizer at rate of 100% NPK alone were insignificantly. Therefore, it could be recommended by using 50% NPK + biofertilizer combined with 400 ppm ascorbic acid to obtain safety food productivity and economically from roselle plants.

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