



Impact of biofertilizers, chicken manure and urea on growth and yield of maize (*Zea Mays* L.) grown in desertified region

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Abstract

In order to assess the effects of biofertilizers and urea (source of chemical nitrogen) and their possible combinations on the performance of a maize variety (Hudeiba 45) in a desert sandy soil, a field experiment was conducted at El Rawakeeb Research Station for two consecutive seasons (2009 and 2010). The field experiment was arranged in a randomized complete block design with four replicates. The treatments used were *Azospirillum brasilense* (A), *Azotobacter sp* (B), chicken manure (CM) at a rate of 7t/ha, urea (46% N) at 40kgN/ha (T), urea at 120kg N/ha (N), A+T, B+T, A+CM and B+CM in addition to and the control. Data were collected after 6, 10 and 14 weeks from sowing on plant height, number of leaves/plant, leaf area, stem diameter, shoot dry weight/plant and root dry weight/plant. Grain yield was also measured. The obtained results showed that chicken manure solely or in combination with biofertilizers significantly ($p \leq 0.05$) increased growth and crop yield. However, in most studied parameters no significant differences were observed between the recommended rate of urea and chicken manure or its combination with the bioferilizr. 33.13% and 24.73% increment in grain yield (t/ha) were recorded when Chicken manure was applied in 2009 and 2010, respectively.

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Introduction

Sudan is one of the most seriously affected countries by desertification in Africa. The arid and semi-arid lands cover an area of more than 90% of the total area of the Sudan (Abdellatif and Elhag, 2015). Since the marked decrease in the productivity of desertified soil is mainly due to low fertility, the use of inorganic fertilizers to increase crop yield is becoming a common practice especially in developing regions. However, high demands and low supply of fertilizers make their cost unaffordable (Toth and Foddor, 2015). Microbial inoculants and organic amendments are cheap natural nutrient inputs that could serve as complements or alternatives to chemical fertilizers for improving crop production in low-input agriculture (Abdullahi *et al.*, 2013)). Many attempts have been made to partially replace these fertilizers by safe and cost effective biofertilizers in maize cultivation (El-Kholy *et al.*, 2005). *Azospirillum* and *Azotobacter* are diazotrophs that fix nitrogen as free living organisms and are used as biofertilizers for many crops. Inoculation of maize seeds with *Azospirillum* and *Azotobacter* was found to increase plant growth, nutrient uptake and yield (Dobbelaere *et al.*, 2001; Kouchebagh *et al.*, 2012). Integrated management strategies involving inoculation of seeds with *Azotobacter sp* and *Azospirillum sp* in combination with chemical fertilizers were found to improve both growth and yield of crops (Ibrahim *et al.*, 2015). Combining chemical fertilizers with biofertilizers is considered as one of the possible solution to minimize the risk of accumulation of chemical fertilizers in the environment (Rueda *et al.*, 2016).

Application of organic manure to sandy soils plays an important role in improving soil media through modifying soil physical and chemical properties (Badawy, 2008). Beneficial effects of organic manure such as increasing maize grain yield and its quality, enhancing soil water retention and decreasing the loss of soil moisture were all reported by Bakry *et al.* (2009). The role of organic fertilizers alone or in combination with biofertilizers has recently gained recognition in sustainable crop production (Abdullahi *et al.*, 2013). Millet production in low-input

agriculture was improved by application of biofertilizer and organic manure either singly or in combination (Abdullahi *et al.*, 2014).

Since 1992, the National Centre for Research in Sudan has conducted applied research to combat desertification at ELR awakeeb Research Station (lies in desertified, tropical semiarid area). Restoration of soil fertility by application of biofertilizers is one of the soil reclamation projects that are carried out to control desertification. Moreover, the available reports of the importance of using biofertilizers for increasing maize growth and yield in desertified area in Sudan are scarce. Considering this, the present study was carried out to develop cost effective and ecofriendly sustainable system where the supply of nutrients can't be ensured. The effects of *Azospirillum brasilense*, *Azotobacter sp*, chicken manure and urea alone or in combination on growth and yield of maize plant grown in desertified area were also studied.

Materials and methods

Sources of Seeds and Inoculants

A maize variety (Hudeiba 45) was provided by the Agricultural Research Corporation in Sudan. The effects of chemical, organic and biological fertilizers on the performance of maize variety were evaluated. *Azospirillum brasilense* and *Azotobacter sp* inoculants were obtained from Biofertilization Department, Environment and Natural Resources Research Institute, National Centre for Research, Sudan.

Site

The field experiment was conducted at the Farm of El Rawakeeb Research Station, National Centre for Research, southwest of Khartoum (Latitude 15° 30' N., Longitude 32° 10' E. and 420 meters above the sea level) for two consecutive seasons. El Rawakeeb climate is characterized by a short rainy season (July – October) and high evaporation potential. The relative humidity is low (9 – 20%) which indicates the arid nature of the area. Air temperature values fluctuate and show marked rise in May (47°C) and

drop in July and August (32°C) due to the incidence of rains (Abdellatif, 2003). Soil was analyzed according to the standard methods described by Richard (1954) and Anderson and Ingram (1993).

Experimental Design

The field experiment was laid out in a randomized complete block design with four replicates. The treatments used were *Azospirillum brasilense* (A), *Azotobacter sp* (B), chicken manure (CM) at a rate of 7t/ha and nitrogen (N) in the form of urea (46% N) as a source of chemical nitrogen was applied at two rates: 120kgN/ha (N) and 40 kNg/ha (T), *Azospirillum brasilense* with urea (A+T), *Azotobacter sp* combined with urea (B+T), A+CM and B+CM in addition to the control.

Land Preparation

The land was disc ploughed, harrowed, leveled then ridged. The area was divided into 4×4 m² plots with 5 ridges 70 cm apart. Four seeds were sown per hole with 20cm between holes. Sowing was carried out in the last week of October in 2009 and the first week of November in 2010. Maize seeds were coated by biofertilizers using 40% gum arabic solution. Chicken manure was provided by the Animal Research Centre, Sudan, and applied at a rate of 7 t/ha. It was distributed on the ridged-plot and mixed with soil. The plots were then irrigated twice for two weeks

before sowing. The soil was irrigated immediately after sowing and at seven days intervals. Plants were thinned manually into one plant/hole after a week from sowing.

Data Collection

Data were recorded using a sample of six plants taken randomly from the outer two ridges in each plot. Samples were collected after 6, 10 and 14 weeks from sowing and parameters such as plant height, number of leaves/plant, leaf area, stem diameter, dry weight/plant (shoot and root) and grain yield were determined. Nitrogen content was determined using Kjeldahl method according to Anderson and Ingram (1993). Minerals were measured by Atomic Absorption Spectrophotometer apparatus.

Statistical Analysis

Data were subjected to standard statistical analysis and means were separated for significance using Duncan Multiple Range Test (DMRT) at 5% level of significance (Gomez and Gomez, 1984).

Results and discussion

El Rawakeeb soil is found to be sandy clay-loam, slightly alkaline (pH 7.5), very poor in nitrogen and phosphorus with adequate content of the exchangeable potassium (Table 1).

Table 1. Some physical and chemical properties of Elrawakeeb soil.

Particle size distribution (%)			Total	Total	Total N	EC	pH
clay	silt	sand	K (Cmol/kg)	P (ppm)	%	dS/m	(paste)
29	22	49	0.48	1.77	0.0144	1.35	7.5

Plant height

Application of the studied treatments showed significant ($P < 0.05$) increase in plant height in most treatments except A and B (Table 2). In the first season, the highest plant height was obtained in plots treated with B+CM at all sampling times. The percentages of increment were 14 %, 12 % and 13.9 % at 6,10 and 14 weeks after sowing compared to the control, respectively. In the second season, the treatment CM showed the highest plant height in all sampling times, giving 16.9 %, 17.5 % and 13 %

increase at 6, 10, and 14 weeks after sowing compared to the control, respectively.

Several studies indicated an increase in plant height as a result of organic and biofertilizers application (Boateng *et al.*, 2006). In the present study, higher plant heights were recorded under chicken manure treatments solely or in combination with *Azotobacter sp*.

This increment in plant height could be attributed to the fact that organic manure represent a good energy source for *Azotobacter sp* while in turn grantee the availability of nitrogen and growth promoting hormones to the crop (Abdullahi *et al.*, 2014).

Application of *Azotobacter sp* combined with 40kg N /ha, improved plant height especially at 14 weeks from sowing and produced results comparable to those obtained when nitrogen was applied at 120kgN/ha. The combined application of

Azospirillum brasilense and 60% N produced comparable results as to those obtained due to the application of the recommended doses of fertilizers (Yadav *et al.*, 2011).

It is well known that the chemical fertilizers promote plant growth through the role of nitrogen in protein synthesis and increasing the meristmatic activity.

Table 2. Effect of applied treatments on plant height/maize plant.

Treatment	First season			Second season		
	Weeks after sowing			Weeks after sowing		
	6	10	14	6	10	14
Control	53.21cd	103.75ef	142.00d	68.00de	118.00d	149.75d
N	52.41d	107.28cde	154.50abc	72.75bcde	130.50bc	165.75a
A	50.79d	101.50f	141.00d	71.25cde	125.50c	153.50cd
B	51.04d	104.25def	143.50cd	67.50e	124.25cd	154.25bcd
CM	57.95ab	112.75ab	156.75ab	79.50a	138.74a	169.50a
A+CM	54.95bcd	110.75bc	153.83abc	73.75abcd	133.75ab	165.25a
B+CM	60.67a	117.13a	161.75a	77.00abc	130.50bc	166.00a
A+T	52.04d	108.75bcd	149.00bcd	68.75de	128.75bc	160.25abc
B+T	56.89abc	113.50ab	158.00ab	77.75ab	133.75ab	163.75ab
mean	54.72b	108.77b	151.15a	72.92b	129.39b	160.89b
C.V%	4.59	3.02	5.25	5.63	3.71	4.16

* Within each column, means have the same letter(s) are not significantly different according to DMRT at 5% level of significance. The means of overall means of each stage were compared together.

Number of leaves per plant

Individual analysis of variance revealed highly significant differences ($p < 0.01$) among the

undertaken treatments for the number of leaves in both seasons and at the three stages of growth (Table 3).

Table 3. Effect of applied treatments on number of leaves/ maize plant.

Treatments	First season			Second season		
	Weeks after sowing			Weeks after sowing		
	6	10	14	6	10	14
Control	3.25bc	6.50b	10.00c	2.50c	5.00c	7.75c
N	3.50abc	6.25b	10.00c	4.00b	6.50b	9.25ab
A	2.75c	5.75b	9.50c	3.50b	6.25b	8.25bc
B	2.75c	6.00b	10.25bc	3.50b	6.00b	8.50bc
CM	4.25a	7.50a	11.25ab	5.00a	7.75a	10.25a
A+CM	3.50abc	6.50b	10.25bc	3.50b	6.75b	9.25ab
B+CM	4.00ab	7.75a	11.75a	4.00b	6.75b	9.25ab
A+T	3.00bc	6.00b	9.50c	5.00a	8.00a	10.00a
B+T	3.50abc	6.50b	10.50bc	5.25a	7.75a	10.25a
Overall mean	3.39b	6.53b	10.33a	4.03b	6.75b	9.19b
C.V%	18.29	9.64	7.90	15.11	9.67	10.29

17.5 % and 32 % increase in the number of leaves per plant at 14 weeks after sowing were recorded for B+CM and CM in first and second seasons, respectively. 32 % increases in number of leaves per plant were also recorded at 14 weeks after sowing for B+T in the second season. Mostly, throughout the different sampling times and during the two seasons, the highest number of leaves was recorded for CM alone or in combination with the bacterial inoculants.

An increase in leaf number in this study could be attributed to the relatively higher availability of nutrients in CM which promoted vigorous foliage growth, increased meristematic activity and intensified physiological activities in plants and hence favored the synthesis of more photo assimilates and early flowering. These results are in consistent with those of Efthimiadou *et al.*, (2009) and Uwah *et al.*, (2011).

Table 4. Effect of applied treatments on stem diameter (cm) of maize.

Treatments	First season			Second season		
	Weeks after sowing			Weeks after sowing		
	6	10	14	6	10	14
Control	0.69b	1.13d	1.53c	0.66a	1.14abcd	1.55cde
N	0.75ab	1.23abc	1.61ab	0.67a	1.17abc	1.59bcd
A	0.70b	1.14d	1.53c	0.60a	1.10cd	1.52de
B	0.74ab	1.20bcd	1.57bc	0.64a	1.11bcd	1.53de
CM	0.78a	1.26ab	1.63ab	0.69a	1.24a	1.71a
A+CM	0.71b	1.23abc	1.62ab	0.67a	1.21a	1.65ab
B+CM	0.78a	1.29a	1.64a	0.67a	1.23a	1.63abc
A+T	0.70b	1.16cd	1.54c	0.64a	1.17abc	1.59bcde
B+T	0.72ab	1.25ab	1.66a	0.65a	1.06d	1.50e
Overall mean	0.73b	1.2b	1.59a	0.64b	1.16b	1.58b
C.V%	5.76	4.27	3.07	12.64	5.84	3.98

Stem diameter (cm)

Stem diameter usually reflects the strength of the plants to resist the stresses. Stem diameter was significantly affected by most treatments in all stages of growth in the first season. The highest stem diameter was recorded for B+CM at 6 and 10 weeks after sowing while the highest increase was recorded for B + T at 14 weeks after sowing. In the second season, stem diameter steadily increased in all plots

treated with CM compared to the control indicating that CM treatment generally enhanced plant growth (Table 4). Similar results were obtained previously by Akongwubel *et al.*, (2012).

Highest stem diameter was recorded when maize seeds were inoculated with *Azotobacter*, *Azospirillum brasilense* and double inoculation of them compared to non-inoculated plants (Hoshang *et al.*, 2011).

Table 5. Effect of applied treatments on leaf area (cm²) of maize.

Treatments	First season		Second season	
	Weeks after sowing		Weeks after sowing	
	6	10	6	10
Control	176.89a	220.00de	169.25a	239.75d
N	173.19a	231.13abcd	195.00a	255.75ab
A	170.70a	216.00e	182.00a	243.50cd
B	176.49a	224.28cde	188.50a	241.70cd
CM	175.00a	240.5ab	198.25a	259.50a
A+CM	173.50a	238.95abc	187.75a	255.25ab
B+CM	181.00a	246.00a	190.00a	256.00ab
A+T	174.00a	230.25bcde	192.00a	252.25abc
B+T	180.33a	241.18ab	200.00a	247.00bcd
Overall mean	175.68b	232.03b	189.19b	250.08b
C.V%	7.90	4.41	10.95	3.02

Leaf area/plant

The influence of the studied treatments on leaf area of maize was presented in Table (5). In comparison with the control, non-significant differences were observed among the undertaken treatments after 6 weeks from

sowing in both years. However, significant differences were recorded after ten weeks in both first and second years. B+CM and CM showed the highest values and significant increase by 11.8% and 8% compared to control in first and second seasons, respectively.

Table 6. Effect of applied treatments on dry weight (g) of maize plant.

Treatments	First season			Second season		
	Weeks after sowing			Weeks after sowing		
	6	10	14	6	10	14
Control	44.58d	102.42cd	142.08b	52.37a	103.63d	139.00d
N	45.46d	104.01bcd	143.01b	57.50a	109.25bcd	145.50abcd
A	46.18cd	104.26bcd	144.26b	56.49a	106.75cd	137.75d
B	50.99bc	100.50d	140.50b	54.50a	104.00cd	139.50d
CM	55.88ab	106.02bc	145.22b	59.63a	115.75ab	151.50abc
A+CM	53.18ab	104.03bcd	143.70b	57.50a	119.52a	153.50ab
B+CM	55.29ab	108.62ab	146.25ab	60.33a	118.25a	154.25a
A+T	55.63ab	103.89bcd	143.70b	57.50a	110.50bc	142.25cd
B+T	56.5a	111.34a	151.59a	58.25a	108.75cd	144.50bcd
mean	51.52b	105.01b	144.38b	57.86b	110.71b	145.31b
C.V%	6.90	3.34	2.95	7.68	4.24	4.53

These results are in agreement with those reported by Belay *et al.* (2001) who stated that improving plant growth by application of organic manure may be attributed to improving mineral nutrition of plants. Organic manures also improve the water holding capacity of the soil, soil structure and aeration. Similar findings were reported also by Amanullah *et al.*, (2009).

Dry weight per plant (g)

Application of the studied treatments gave significant increment ($p < 0.05$) in maize dry weight (shoot and root) per plant in all stages of growth. Particularly, In the first season, the results indicated that inoculated plants with B+T showed higher dry weight as compared with the those receiving the recommended nitrogen rate (120kg N/ha). In the second season, the combined application of B with CM gave the highest values of dry weight (Table 6). The observed significant performance of maize dry weight when *Azotobacter sp* with CM we applied could be attributed to the promoting effect of *Azotobacter sp* on root growth which in turn enhancing nutrients and water uptake (El-Koly *et al.*,

2005). Beside, CM contained essential nutrient elements that increased photosynthesis efficiency and hence enhanced maize dry weight. Our results are in agreement with those obtained for maize (Gholami *et al.*, 2009), wheat (Galal *et al.*, 2000) and sunflower (Ismail and Hasabo, 2000).

Grain yield

Table (7) demonstrates the effect of the various tested treatments on maize grain yield. Highly significant differences were recorded among the treatments for the grain yield per plant and per hectare. Grain yield/plant of the individual treatments means indicated was increased significantly by most of the treatments used compared to control. Application of CM showed significant increment in grain yield in both seasons compared to control. The percentages increment of the grain yield (t/ha) due to application of CM were 33.13% and 24.73% in the first and second season compared to control, respectively. Unexpectedly, no significant differences were observed between the sole application of CM and its combinations with the biofertilizers. Also, application of *Azotobacter sp* with 40 kgN/ha (B+T) did not show

significant difference in yield compared to the application of N (120kgN/ha) in the second season and the percentages of increments were 18.81% and 18.27% for B+T and N, respectively. This means application of B+T saved about 80kgN/ha /season. Our results are in accordance with those obtained by

Ibrahim *et al* (2015), Abdullahi *et al* (2014), Amujoyegbe *et al.* (2007). Also, Rizwan *et al.* (2008) reported that application of chemical fertilizers combined with biofertilizers produced highest grain yield in comparison to the sole application of each of them.

Table 7. Effect of applied treatments on grain yield of maize.

Treatments	First season		Second season	
	Grain yield/plant (g)	Grain yield (t/ha)	Grain yield/plant (g)	Grain yield (t/ha)
Control	21.00e	1.66c	25.75c	1.86e
N	25.46bcd	2.04ab	28.58ab	2.20ab
A	22.67de	1.77c	24.50c	1.93de
B	23.50cde	1.84bc	26.25bc	2.03cd
CM	29.98a	2.21a	31.07a	2.32a
A+CM	26.67abc	2.13a	29.75a	2.24a
B+CM	27.87ab	2.17a	29.05ab	2.17abc
A+T	23.34cde	1.81bc	26.38bc	2.08bcd
B+T	26.83abc	2.11a	29.28ab	2.21ab
Overall mean	<u>25.26b</u>	1.97b	<u>27.76b</u>	2.12b
C.V%	10.79	9.25	7.91	4.89

As observed in this study, application of chicken manure alone or in combination with *Azotobacter sp* and *Azospirillum brazielense* enhanced the plant growth and yield. This could be attributed to the positive effects of chicken manure in decreasing the loss of soil moisture, increasing soil water retention and soil bio-availability of micronutrients (Bavariani *et al.*, 2016). In addition, *Azospirillum brazielense* and *Azotobacter sp* promoted maize growth and yield by their capacity to fix atmospheric nitrogen, produce plant growth regulators (Dobbelaere *et al.*, 2003), enhance plant nutrients availability and drought resistance (Abdullahi *et al.*, 2014).

Conclusion

From the findings of this research, it could be concluded that application of chicken manure and biofertilizers both solely or in combination could improve maize growth and yield in arid land. Results also showed that application of *Azotobacter sp* with 40kgN/ha can save about 80kg N/ha/season

compared to the recommended dose of nitrogen .However; further study might be required for more confirmation.

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