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Effect of Vital Inoculations and Different Nitrogen Forms Fertilizer on the Quality and Productivity of Sunflower Plant under New Valley Conditions

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THIS RESEARCH work was carried out at the new valley area, which is characterized by hot atmospheric conditions. In other words, plants cultivated at that area will definitely need help to overcome the adverse conditions prevailing in that experimental area. Fertilizer management is believed to be a good vehicle for providing sufficient nutrition to the grown plants. Encompassing combinations of mineral N fertilizer forms and free-living atmospheric N fixer organisms, fertilizer management is thought of to be a helpful tool to achieve remedial target anticipated from plants grown under the conditions of the experimental area. The combination treatment composed of applying ammonium sulfate (AS) at 60kg N/fed and inoculating with bio-fertilizer mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* was found to be significantly the best fertilizer management regime for the study area based on the findings of the current work. This combination treatment could produce the highest seed and oil yields of sunflower plants when compared with other combination treatments in both seasons of study. Fertilizer management regime in the current work depicted experimenting with combinations of mineral N forms applied at two rates and combined with inoculation with *Azotobacter chroococcum, Azospirillum lipoferum* and their mixture. The objective of the current research work is mainly to figure out a fertilizer combination treatment to help sunflower plants overcome the harsh conditions prevailing in the study area.

Keywords: Bio-fertilizers, Microbial activity, Nitrogen fertilizers, Sunflower, Yield components.

Introduction

Sunflower (*Helianthus annuus L.*) seeds contain 38-53% oil which is characterized by high content of unsaturated fatty acids and very low content of cholesterol (Abdel-Motagally & Osman, 2010). Nitrogen is the top most important needed nutrient that plays a major role in enlarging crop yield in agriculture. Therefore, nitrogen fertilizers such as ammonium sulfate (AS), ammonium nitrate (AN), and urea have been extensively used in modern agriculture. These fertilizers are generally oxidized to nitrate via nitrite by nitrifying microorganisms in the agricultural field (Isobe & Ohte, 2014). As N enhances yield components and yield of crops, mineral nitrogen forms of ammonium sulfate (AS) and ammonium nitrate (AN) were developed to help attain higher yields of world food, then an organic nitrogen form of urea was recruited to help achieve the same purpose. AS is highly soluble and can be easily vulnerable to leaching with the soil water percolating down the rhizosphere and out of the reach of plant roots if applied in excess to plant needs or when irrigation water is applied in surplus to soil holding capacity. In salt affected soils especially with high pH and high degrees of temperature, ammonium is liable to volatility. Therefore, AS is prone to low use efficiency nitrogen fertilizer.

With regard to nitrate-N fertilizer, besides being highly soluble as compared with AS, it contaminates ground and surface water due to

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nitrate leaching and loss from the agricultural field through de-nitrification by soil anaerobic bacteria (Ju et. al., 2009). Contamination due to nitrate escaping to drainage system and ground water is actually serious where it kills living organisms with especial reference to humans where a concentration of 10mg nitrate/liter was reported to be highly toxic to human and cause intestinal troubles and may lead to death. Lucas & Vitosh (1984) ranked mineral fertilizers according to their salt index, which refers to the damage they could cause to plants relative to that by an equal weight of sodium nitrate. AS, AN, CN and UN had salt index values of 0.74, 1.05, 0.65 and 0.75, respectively. It can be seen that AN proved to be the strongest in harming the grown plants when compared with AS and urea.

Generally, volatility of prices, transportation, and storage costs of chemical fertilizers and endangering human health imposed the use of manure for soil fertility which improves soil fertility, increases water-holding capacity, decreases soil erosion, improves amount of oxygen, and promotes beneficial organisms and productivity (Hamza & Abd-Elhady, 2010). Munir et al. (2007) found that highest seed oil content and oil yield were produced from sunflower plants which received 30kg N/fed of AS, AN, and urea applied either alone or in combination with 20 or 30m^3 farmyard manure. In other words, the application of organic manure may help stop fertilizer losses through leaching. In addition, short irrigation periods at volumes that are just enough to satisfy plant needs of water absorption. Such amounts of irrigation water leaves no extra water to move below the rhizosphere.

FAO (1991) reported that urea could not provide furrow cultivated plants with readily available nitrogen when compared with AS, AN, or CN. This was referred to the chemical and biological reactions that urea had to undergo in the soil, as well as to its movement in the soil profile down below the root zone. While, Nicoulaud & Bloom (1996) concluded that urea can be used to supply part of the N required for plant growth and that failure of urea to promote rapid growth is probably due to its phyto-toxicity and not to N deprivation. This was also referred to that AS resulted in a significant increase in soil acidity (lower pH) compared with ammonium nitrate (AN) or urea.

To avoid the troubles occurring from the application of AS, AN, and free urea, application of slow release fertilizers deemed to be necessary. Urea was coated with various substances amongst which is formaldehyde to protect the urea molecule from the attack of microbial populations existing in the rhizosphere and enable a slow release of nitrogen into the soil during the crop-growing season. The use of controlled-release nitrogen fertilizers is mainly based on the principle of nitrogen utilization efficiency. Such efficiency will definitely decrease in the case of urea coated with formaldehyde owing to elongating the time required by microbial population to hydrolyze the coating then reach the urea molecule and start to release N from it. The delay of N release will not cope with plant needs especially during fast growth stages.

With respect to bio-fertilizers, they contain useful microorganisms, which could colonize in the rhizosphere and promote plant growth through increasing the supply or availability of essential nutrients to plants (Vessey, 2003). Mehran et al. (2011) indicated that manure significantly affected grain yield ($P \leq 0.01$); the highest grain yield was achieved in the interaction of manure × *Azotobacter* × *Pseudomonas* (4.556ton sunflower seeds/ha). They also added that seed yield was not significantly affected by the microorganisms when added alone. Abd El-Rahman et al. (2016) partially substituted the recommended mineral N rate of 30kg N/fed with organic N fertilizer as compost combined with biofertilizer. Their results revealed that application of 15kg N/fed +25% compost +Bio significantly improved yield and yield attributes and protein and oil yields as compared to 100% compost or 30kg N/fed alone. Integrating mineral-N with 25-50% compost always gave higher values than those obtained with full mineral or organic applied separately. They also found that highest seed oil percentage was recorded when plants were fed 100% compost + Bio treatment followed by feeding with 7.5kg N/fed +75% compost. Highest seed oil yield was recorded when feeding with 15kg N/fed + 50% compost followed by 15kg N/fed +25% compost +Bio. Their study points to the need for integration among mineral nitrogen, organic fertilizer, and biofertilizer to attain the highest possible sunflower seed and oil yields. The biofertilizer applied in their work contained 10^8CFU ml^{-1} from each bacterium of *Azotobacter* and *Azospirillum*. This treatment points to the

benefits of coexistence of both microorganisms together and saving considerable amounts of mineral nitrogen.

Kargar et al. (2014) stated that about 90% of nitrogen fixation is performed by microorganisms. In addition, *Azotobacters* are important free-living nitrogen-fixing bacteria. They investigated the role of *Azotobacter chrococcum* and *Azotobacter paspali* in the growth and productivity of wheat. They isolated the most active *Azotobacter chrococcum* and *Azotobacter paspali* in fixation of nitrogen in different wheat fields using *Winogradskii* culture medium. There randomized study was performed in six replications at outdoors considering the following factors; urea at three levels (no urea, 100 mg urea per kg soil, and 200 mg urea per kg soil), and 3- bacteria type at four levels (no bacteria, *Azotobacter paspali* alone, *Azotobacter chrococcum* alone, and a mix of both bacteria). Findings: The highest wheat response was obtained with *Azotobacter chrococcum and Azotobacter paspali*. They also found that the difference between various levels of nitrogen fertilizer was significant at 5% level. They suggested that the studied region's indigenous *Azotobacter chrococcum* enhanced more wheat growth through nitrogen fixation compared to the other *Azotobacter* due to calcareous soil and warm and dry weather of the Province. They also stated that organic compounds such as animal manure degrades gradually and enhances the growth and stimulate nitrogen fixation by free-living bacteria such as *Azotobacters*. The indigenous *Azotobacter chrococcum* can also be isolated and used for biologic fertilizers to improve crop yield.

From another stand point, Herrera et al. (2016) reviewed knowledge about technologies for N fertilization with potential to increase N use efficiency and reduce its negative effects on the environment. They stated that classic inorganic sources such as urea and ammonium sulfate were the major sources utilized, while controlled N release fertilizers have not been significantly adopted for cereals and oil crops. They pointed to that microorganisms, with the exception of *Rhizobium* sp. in soybeans, are also not widely used those days (e.g., plant growth-promoting bacteria and *Cynobacteria*). The interest in implementing new N fertilization knowledge is stimulating the development of sensors to diagnose the N status and decision support systems for integrating several variables to optimize sources, rates and methods of application. Furthermore, increasing concern about the environmental consequences of N may facilitate the implementation of innovations outside the farm such as more effective regulations to guide N fertilization and methods to manufacture N fertilizers that are more energy-efficient and less CO_2 equivalent emitting.

Gendy et al. (2013) carried out a field experiment during two successive seasons, 2011 and 2012 aiming to study the effect of utilizing different sources of nitrogen (ammonium nitrate NH4NO3 or ammonium sulphate $(NH_4)2SO_4$ with or without adding bio-fertilizers (biogein at 2kg/fed., nitrobein at 2 kg /fed., or biogen at 1kg/fed., + nitrobein at 1kg /fed.) as well as their interaction on the plant growth, seed yield, total protein and total guaran content in seeds as well as some chemical contents in leaves. Their results revealed that different sources of nitrogen or bio-fertilizers increased the growth parameters; i.e., plant height, number of branches per plant and dry weight of aerial part and leaves per plant, as well as number of pods/plant, weight of seeds (gm/ plant or kg/ fad.), and chemical constituents such as total protein and N, P, K compared to untreated plants. They stated that fertilizing guar plants with ammonium sulphate was the most effective in raising the productivity of seeds and the content and yield of guaran and chemical composition than ammonium nitrate. Treating plants by bio-fertilizer (mixture of biogein+nitrobein) was the most effective in this concern followed by nitrobein and then biogein. The interaction treatment of ammonium sulphate at 60kg N/fed., + bio-fertilizer (biogein at 1 kg/ fed., + nitrobein at 1kg/fed.) gave the best result in this concern with significant differences if compared to the control and the other treatments under study in both seasons.

The aim of this study is to investigate the integrated effects of mineral-N fertilizers; ammonium sulfate and ammonium nitrate, and that of organic-N urea-formaldehyde all applied at two rates without or with the inoculation with bio-fertilizers; *Azotobacter chroococcum* and *Azospirillum lipoferum* and both together, on sunflower seed and oil yields, and quality, as well.

Materials and Methods

Agricultural experiment

A field experiment was conducted in a sandy soil at El-Monira village, El-Kharga Oasis of the GIS indices of 30.53 longitude, 25.45 latitude and 78.8m latitude at the New Valley Governorate during two successive cropping seasons of 2016 and 2017. The major objective of the current research was to study the response of sunflower plants to mineral nitrogen fertilizers; ammonium sulfate (AS), ammonium nitrate AN), and urea formaldehyde (U), applied at two levels; 60 and 30kg N/feddan (1 feddan= 0.42hactar). All of the previous six combination treatments were applied without (control) and with being inoculated by *Azotobacter chroococcum*, *Azospirillum lipoferum*, and their mixtures.

To apply the adopted microorganisms, bacterial concentration of the applied suspensions was adjusted to 10^8 CFU/ml for the three microbial treatments. Sunflower seeds were wetted before planting with bacterial suspensions at the rate of 250ml suspension/1250gm seeds for three hours before planting. Carboxy methyl cellulose 0.5% was used as an adhesive agent; i.e. spreading agent. The previous pre-planting treatment with microbial suspensions represents half dose of the biofertilizer treatments. The other half was applied in the form of spray using a back-mounted sprayer to soil down plants aging one month after seeding.

With regard to mineral fertilizers, they were applied in three doses; after 30 (at vigorous vegetative growth and plant elongation growth stage), 45 (at flowering), and 60 days (at head maturity and seed filling growth stage) from seeding. The three mineral nitrogen fertilizers; AS, AN, and U-formaldehyde, were applied in the form of solid fertilizer about 7cm away from plant stem to avoid harming those plants, except for urea formaldehyde which was applied immediately down the plants close to the stems owing to the slow solubility of that fertilizer and expecting no harm to the plants. The three application doses were not equal, with the first being the least because the plants were not in big need for N nutrient at that dose. The second dose of fertilizer N was the greatest because it coped with the fast growth time. The third dose coincided with the heading growth stage which witness pollination and seed production.

With regard to phosphorus fertilizer application, it was applied at the rate of 45kg P/fed with the applied organic manure which was added at the rate $20m^3$/feddan 15 days prior to seeding. The applied amounts were metered per dripper line and were to be applied in the furrow that was manually dug, then the furrow was filled with the dugout soil. This practice was achieved the same for all treatments.

With respect to potassium fertilizer, it was applied the same to all combination treatments in all replicates in two equal doses after 45 and 60 days from seeding. The applied dose was to be calibrated per plant and applied 5-7cm away from plants stems to avoid plant burn damage.

Farm yard manure was broadcast applied and incorporated into the soil surface layer by hand hoeing, then the dripper lines were straight back to their places. A quick false irrigation was applied on to the experimental dripper lines through the drip irrigation network 15 days before seeding to allow the exposure of weeds that may be existing in the soil and the added farm manure to combat them.

Seeding rate was 5kg/fed. On the seeding day, 3-4 seeds were to be placed in hills 30cm apart along the dripper lines. After germination, number of plants was thinned to 2 plants per hill to achieve a full 100% stand at the beginning of the cultivation season. Owing to the fine textured soil nature (around 48% as silt + clay), drip irrigation was to be practiced every six-day period with short application period each time to avoid loss of soluble fertilizers with the soil water percolating down the rhizosphere, i.e. beyond the plants root system.

Some physical and chemical analyses of both the adopted soil and irrigated water:

Some physical and chemical analyses of the experimental field soil were achieved according to Page et al. (1982) and presented in Table 1. In addition, chemical analyses of irrigation water were depicted in the same table. The latter analyses exhibited that the irrigation water had pH value, 7.5 and EC, 1.46 $dS.m^{-1}$.

Test plant

Seeds of the adopted test plant (Sunflower cultivar Sakha 53 (*Helianthus annuus* L.) were purchased from the Oil Crops Research Institute, Agricultural Research Center, Giza, Egypt.

TABLE 1. Analysis of experimental soil to a 30cm depth and irrigation water

Physical analysis %			Soil texture
Sand	Silt	Clay	
52.95	21.51	25.54	Sandy clay loam

Chemical analysis												
pH	E.C dS.m^{-1}	$CaCO_3$ %	O.M %	T.N %	Cations meq/L				Anions meq/L			
					Ca^{+2}	Mg^{+2}	Na^{+}	K^{+}	CO_3^{-2}	HCO_3^{-}	Cl^{-}	SO_4^{-2}
8.64	8.21	4.34	0.58	0.09	25.2	8.2	43.4	5.3	0.9	15.9	50.9	14.4

Chemical analysis of irrigation water										
pH	E.C dS.m^{-1}	Ca^{+2}	Mg^{+2}	Na^{+}	K^{+}	CO_3^{-2}	HCO_3^{-}	Cl^{-}	SO_4^{-2}
7.5	1.46	5.42	3.24	5.43	0.27	1.65	2.18	4.2	6.16

Microorganisms

Bacterial isolates recruited in the current experiment were isolated from salt affected soils at newly reclaimed sandy areas. The isolated bacteria were identified as being *Azotobacter chroococcum* and *Azospirillum lipoferum* according to Bergey's Systematic Bacteriology (Krieg & Dobereiner, 1984).

Plant growth promoting properties of bacterial isolates: Agronomic data recoded:

Soil biological activity

Enumeration of microorganisms in soil samples were carried out by the most probable number (MPN) technique. One milliliter successive dilutions of 10^3, 10^4, and 10^5 were attained. Soil samples were to be transferred to test tubes containing semi-solid NFb medium and plates of Ashby medium for *Azospirillum* and *Azotobacter* isolates, respectively. Tubes and plates were then incubated under suitable temperature. Microorganisms were identified based on cultural, morphological and biochemical characteristics as per Bergey's Manual of Systematic Bacteriology.

Bacterial population = (MPN value × middle dilution × middle dilution used)/ (Dry weight of the soil sample)

Determination of microbial activity

Counts of microorganisms were estimated by the dilution plate technique methods. The following microbial analyses; total *Azotobacter* count (Counts x 10^4CFU g^{-1} dry soil) and *Azospirilla* densities (Counts x 10^4CFU g^{-1} dry soil) were carried out in all soil samples. Dehydrogenase activity (µg TPF g^{-1} dry soil 24hrs) in the rhizosphere soil was determined according to Pramer & Schmidt (1964) and Thalmann (1967). Production of ammonia was determined according to Cappuccino & Sherman (1992).

Soil analyses

Soil samples were collected from each dripper line at the same time of plant sampling, air-dried, passed through a 2 mm sieve and kept for physical and chemical analyses. Particle size distribution was determined using the pipette method according to Jackson (1973). Electrical conductivity (EC) and soil pH was determined in a 1: 2.5 soil to water extract using conductivity meter and Beckman pH meter, respectively according to Jackson (1973). Organic carbon content was determined by Walkley and Black's wet oxidation method (Walkley & Black, 1934). Available potassium was extracted by neutral normal ammonium acetate method and measured by flame photometer. Available P was extracted using 0.5 M $NaHCO_3$ at pH 8.5 according to Olsen et al. (1982) and measured colorimetrically using the chlorostannus phosphomolybdic-sulfuric acid method as described by Jackson (1973).

Chemical composition of plants and seeds:

Half gram of the powdered plant samples was digested using tertiary acid mixture ($HClO_4$ + HNO_3 + H_2SO_4) according to Jackson (1973). Phosphorus was determined using a spectrophotometer, but K by a flame photometer according to Jackson (1973). Another half-gram was digested by the modified Kjeldahl method for N determination according to Jackson (1973).

Yield and yield components

At harvest, plant height (cm), number of leaves/ plant, seed head fresh weight (g), seed head dry weight (g), plant weight (g), seeds weight (g)/plant, and seed head diameter (cm), then seed yield (kg/ feddan) (1 feddan= 0.42hactare) was calculated in both seasons of the current study. In addition, oil production (g/plant) was also assessed and calculated as kg/feddan. To evaluate effects of the applied treatments on seed quality, seed mineral content of N, P, and K was achieved.

Soil physical and chemical properties of the experimental site were measured according to Page et al. (1982). Soil texture was achieved by dry sieving analysis. In soil paste extract, soil pH, EC, and water soluble cations and anions were measured. Extracted Na^+ and K^+ were measured flame photometrically, Ca^{++} and Mg^{++} titrimetric ally using versinate, HCO_3^- titrimetric ally using dilute HCl, and Cl^- titrimetric ally using silver nitrate. Sulfate was calculated as the difference between cations and anions. Calcium carbonate content of the soil was achieved using a calcimeter. Organic matter content was also measured according to Page et al. (1982). Available N was measured according to Onken & Sunderman (1977). Available P content was achieved according to Olsen et al. (1982), while available K was measured using ammonium acetate solution according to Page et al. (1982).

Statistical analysis

All dripper lines were divided into three groups of drippers according to the expected drop in the driving head along the end tail of lines. The three groups were to be sampled for three replicates, respectively. The obtained data were exposed to the analysis of variance (ANoVA) according to the statistical design of split-split plot technique in randomized complete blocks using SPSS (2014) software package. The Duncan least significant range (LSR) will be recruited to differentiate among treatment means obtained in the current work. The result of using Duncan test will show up in table cells as alphabets attached close to the right of various mean values.

Results and Discussion

Soil available N, P, and K

Initial soil available nitrogen was found to be 0.008% (80µg/g dry soil) on the average for all soil samples collected at the time of seedbed preparation. This amount of available N resembles the starter dose of N on which the plants and microorganisms were going to start their activities. Doing so gives us the opportunity to follow up the influence of the applied treatments on the adopted test plant sunflower. Based on the analysis results no gross variations were found among the samples collected from the soil under all dripper lines. Therefore, the starter situation is not considered a source of variability.

Data in table 2 show that all applied treatments and the control enhanced soil available nitrogen content at the time of sunflower flowering above the initial content. This can be referred to a changed effect due to the application of fertilizers.

Data in Table 2 show that AS could significantly surpass AN which, in turn, surpassed urea-formaldehyde in proliferating greater available nitrogen in the rhizosphere that accommodates sunflower roots in the first season. With regard to the application rate, the higher application rate significantly proved to be better in bringing more available N. These findings coincide with those obtained by FAO (1991) which pointed to the same order for the three nitrogen fertilizer forms. In addition, data in the same table point to that the interaction between N sources and rate of application was significant. In other words, the three N forms were steadily greater with the higher N rate of application than with the lower one in bringing available N in the rhizosphere.

From Table 2, it is clear that the application of bio-fertilizers proved to be significantly beneficial over the control treatment in accumulating greater available N in the rhizosphere of sunflower plants in the current work. Albeit obvious, *Azotobacter* exceeded *Azospirillum*. But, applying both microbes together was much better than applying both organisms separately. Thus, including *Azospirillum* with *Azotobacter* could foster the effect of *Azotobacter* alone forward to a peak of influence on soil available N in the rhizospher of sunflower plants. These findings agree strongly with what was reported by Mehran et al. (2011) who stressed the advantages of applying both organisms together than being separately applied. Same Table 2 points also to that the interaction between N forms and bio-fertilizer treatments was found significant. The best combination treatment was obtained from applying AS with the mixed bio-fertilizer treatment of *Azotobacter*-*Azospirillum*.

TABLE 2. Soil available N, P, and K at sunflower flowering stage as affected by the applied treatments in 2016 and 2017

N forms	Bio	Soil av. N (µgN/gm soil)				Soil av. P (µgP/gm soil)				Soil av. K (me/100gm soil)			
		2016		2017		2016		2017		2016		2017	
		60	30	60	30	60	30	60	30	60	30	60	30
AS	Control	217 k	184 lm	226 q	196 s	3.12 q	2.66 t	3.72 ij	2.90 p	2.14 l	2.01 m	2.17 lm	2.11 o
	Azotobacter	392 d	328 f	400 f	336 j	4.05 g	3.59 i	4.58 e	3.82 hi	2.31 f	2.20 hi	2.38 g	2.27 hi
	Azospirillum	332 f	278 i	340 i	287 n	3.76 i	3.18 o	4.27 f	3.44 lmn	2.25 g	2.15 jkl	2.30 h	2.19 kl
	Mixed	563 a	422 c	578 a	434 d	5.66 a	4.93 c	5.74 a	5.33 b	2.87 a	2.60 c	2.98 a	2.71 c
AN	Control	200 l	169 m	208 r	176 u	3.00 r	2.21 u	3.42 mn	2.43 q	2.06 m	1.86 n	2.13 mno	2.05 p
	Azotobacter	368 e	297 h	378 h	312 l	3.90 h	3.41 m	4.05 g	3.75 i	2.24 g	2.17 ijk	2.36 g	2.22 jk
	Azospirillum	313 fg	253 j	323 k	261 o	3.71 j	3.18 o	3.93 h	3.34 no	2.18 ij	2.13 l	2.29 hi	2.16 lmn
	Mixed	508 b	388 d	516 b	406 e	5.24 b	4.40 e	5.33 b	4.80 d	2.68 b	2.45 d	2.76 b	2.66 de
Urea-F	Control	186 l	162 n	191 t	166 v	2.68 s	2.11 v	2.96 p	2.28 r	1.89 n	1.80 o	2.06 p	1.90 q
	Azotobacter	329 f	291 hi	338 ij	298 m	3.70 k	3.37 n	3.95 gh	3.53 klm	2.23 gh	2.15 jkl	2.25 ij	2.19 kl
	Azospirillum	302 gh	252 j	311 l	256 p	3.40 m	3.14 p	3.63 jk	3.27 o	2.16 jkl	2.04 m	2.24 ij	2.12 no
	Mixed	490 b	294 h	505 c	386 g	4.91 d	4.25 f	5.05 c	4.54 e	2.65 b	2.37 e	2.63 e	2.53 t

Mean values in the above table sharing one alphabet are not significantly different

From Table 2, it is clear that the application of bio-fertilizers proved to be significantly greater with the higher rate of applying N forms than with the lower one. This normally point to a significant influence of the interaction between bio-fertilizer treatments and rate of application. The best combination treatment was mixed bio-fertilizer with the higher rate of applying N forms.

From Table 2, it is clear that the best combination treatment for accumulating soil available N in the rhizosphere of sunflower plants under the conditions of the current work can be figured out as the application AS at the rate of 60kg/fed accompanied by the application of mixed *Azotobacter* and *Azospirillum* bio-fertilization in both seasons of study season.

Data regarding soil available N in the second season are depicted in Table 2, from which, it can be seen that AS could significantly accumulate higher available N than the other two sources. By the time, the mixed application of the AS proved to be the best technique of N forms application when compared with the sole application of both forms alone. Regarding to the rate of application of N forms, the higher rate proved to be significantly better than the lower rate in proliferating more N in the rhizosphere of sunflower plants.

Table 2 tells also that the mixed bio-fertilizer treatment; *Azotobacter+Azospirillum*, proved to be significantly more active than the application of both *Azotobacter* and *Azospirillum* alone. Albeit clear, *Azotobacter* was better than *Azospirillum* when applied separately. The best treatment is that of applying AS at 60kgN/fed. In addition, same table 2 shows that the best bio-fertilizer treatment is *Azotobacter+Azospirillum* with AS at the 60kg N/fed.

In conclusion, the best applied treatment was figured out to be that combined from ammonium sulfate (AS) + ammonium nitrate (AN) applied at the rate of 60kg N/fed with the inoculation of biological treatment that is composed of a mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* which free-living atmospheric nitrogen fixers.

With regard to effects of the applied treatments on soil available P, data in Table 2 approximately exhibit that the same effects observed on soil available N showed up on soil available P, except for the interaction between N sources and rate of application in the second season was not significant. This means AS was better than AN, which was, in turn better than urea-formaldehyde, irrespective of being applied at either of the two

adopted rates. However, the higher rate proved to be significantly better than the lower one.

Same Table 2 tells also that soil available P responded significantly better with the mixed application of biological fertilizer mixture of *Azotobacter* & *Azospirillum*. Table 2 again tells that the interaction between N sources and bio fertilizer and the interaction between rate of application and bio fertilizer were found significant. Consequently, the best combination treat in effecting highest soil available P in the rhizosphere of sunflower plants is that composed of AS at 60kg N/fed with inoculation with biological fertilizer mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum*.

From Table 2 trends showing up regarding soil available P were, more or less, the same regarding soil available K. the interaction between N sources and rate could not prove to be significant in both seasons of the current study. Consequently, the best combination treat in effecting highest soil available K in the rhizosphere of sunflower plants is that composed of AS at 60kg N/fed with inoculation with biological fertilizer mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum*.

Density of Azotobacter chroococcum and Azospirillum lipoferum

Initial density of total *Azotobacter chroococcum* counts 5.6×10^3CFU/g dry soil at the time of seedbed preparation. While the initial density of total *Azospirillum lipoferum* counts 6.8×10^3CFU/g dry soil at the time of seedbed preparation.

Data in table 3 show the influence of applied treatments on *Azotobacter chroococcum* and *Azospirillum lipoferum* at the flowering growth stage of sunflower plants in the two seasons of the current study.

Table 3 declares that AS was significantly better in effecting for highest *Azotobacter* densities in the rhizosphere of sunflower plants than AN, which, in turn, was significantly better than urea-formaldehyde. This result can be explained by that AS proliferated more soil available NPK in the rhizosphere area. The higher rate of applying N forms proved to be significantly more effective in promoting greater growth of *Azotobacter*. With respect to the biological performance of the two adopted microbes in the current study, it can be seen from table 3 that applying them was more promoting to *Azotobacter* growth in the rhizosphere as compared to the control (un-inoculated) treatment. Inoculation with *Azotobacter* was more promoting of its growth as compared with inoculation with *Azospirillum*. In addition, inoculation with a mixture of both microbes was significantly more effective in promoting *Azotobacter* growth in the rhizosphere.

TABLE 3. *Azotobacter* and azospirilla densities (counts x 10^4CFU/g dry soil) in the rhizospher of sunflower at flowering stage as affected by the applied treatments in 2016 and 2017

N forms	Bio	*Azotobacter* counts x 10^4CFU/gm dry soil				*Azospirilla* counts x 10^4CFU/gm dry soil			
		2016		2017		2016		2017	
		60kg N	30kg N	60kg N	30kg N	60kg N	30kg N	60kg N	30kg N
AS	Control	36 l	34 m	48 q	4 r	4.75 lm	4.62 m	5.47 l	5.13 m
	Azotobacter	70 c	71 c	76 e	79 c	5.87 g	5.45 j	6.98 g	6.70 h
	Azospirillum	65 d	66 d	72 g	74 f	6.58 c	6.33 d	7.62 de	7.30 f
	Mixed	72 b	75 a	94 b	97 a	7.88 a	7.70 b	9.57 a	9.25 b
AN	Control	25 n	22 o	36 s	34 t	3.88 o	3.67 op	4.65 n	4.23 o
	Azotobacter	56 h	52 i	67 j	62 m	5.62 hi	4.72 m	6.07 i	5.75 j
	Azospirillum	51 i	48 jk	64 l	58 n	5.72 h	5.28 k	6.17 i	5.88 j
	Mixed	65 d	60 f	76 de	71 h	6.18 e	5.92 fg	7.95 c	7.73 d
Urea-F	Control	23 o	21 p	29 u	25 v	3.55 p	2.90 q	4.28 o	3.63 p
	Azotobacter	51 i	49 jk	57 n	53 o	4.88 l	4.42 n	5.43 l	5.07 m
	Azospirillum	48 k	47 k	53 o	50 p	5.68 hi	5.20 k	5.80 j	5.40 l
	Mixed	63 e	58 g	69 i	66 k	6.02 f	5.55 ij	7.50 e	6.68 h

Mean values in the above table sharing one alphabet are not significantly different.

The interaction of N forms x rates showed up to be significant, and so did the interaction of N forms x bio-fertilizer could prove to be significant. But, the interaction of rate x bio-fertilizer could not show up as being significant. This result can be explained by that the inoculated microbes could compensate for the difference between the two rates. In other words, the measured microbe *Azotobacter* could utilize the biological variations in the rhizosphere and went ahead to nourish irrespective of the rate of N forms application. The 3-variable interaction was not significant under the conditions of the current study. These findings held true for both seasons of study, except for the 3-variable interaction which was significant in the second season only. After all it can be concluded that the best combination treatment was AS at 60kg/fed with the inoculation with a mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* for proliferating the highest *Azotobacter* counts in the rhizosphere around sunflower plant roots in both seasons.

Data in Table 3 tells that approximately the same trends observed for the influences of the applied treatments on *Azotobacter* growth held true for the growth of *Azospirillum*. Exception were the interaction of rate x bio-fertilizer and that of the 3-variable interaction, which were not significant in the second season only, while they were significant in the first season.

In general, *Azotobacter* total count was much greater than that of *Azospirillum* in both seasons. This result held true even with inoculation with *Azospirillum*. It seems as if *Azotobacter* can take advantage on *Azospirillum* and show more vigorous growth in the rhizosphere of sunflower plants. This result means that this combination treatment possesses the highest potential for fixing atmospheric nitrogen by the free living microbe *Azotobacter*. Logically, this result stands behind the superiority of this combination treatment in accumulating the highest available soil nitrogen in the rhizosphere of sunflower plants as shown in Table 2.

CO_2 evolution and dehydrogenase activity (DHA)

Initial CO_2 evolution was found to be 6.75mg/ 100g soil/ 24hrs, while initial dehydrogenase activity was 31.4mg TPF/ g dry soil/ 24hrs, at the time of preparing the seed before planting. Table 4 depicts the values of CO_2 evolution as mg/ 100gm dry soil/ 24hrs and exhibits the influence of the applied treatments on that process. Recruited N forms can be arranged in the following significant descending order AS > AN> urea-formaldehyde in daily CO_2 evolution in the rhizosphere around sunflower plant roots. The 60kg N/fed was significantly better than the lower 30kg N/fed. The third variable bio-fertilizers showed that inoculation with either *Azotobacter chroococcum* or *Azospirillum lipoferum* alone could significantly exhibit increases above the control treatment in CO_2 evolution. By the time *Azotobacter* surpassed *Azospirillum* in CO_2 evolution when they are applied separately, the mixed application of microbes could perform significantly better. These findings were true in both seasons of study.

The influence of the three 2-variable interactions and the 3-variable interaction could not prove to be significant on CO_2 evolution in both seasons of study, except for the N forms x bio-fertilizers was found significant in the second season only. Such result can be referred to the clear-cut effect of the sole variables levels on CO_2 evolution in the rhizosphere under sunflower plants. In general, this superiority points to the highest energy that is exerted in the rhizosphere by the eruptions of microbial growth.

In conclusion, the best combination treatment was that composed of applying ammonium sulfate (AS) at the rate of 60kg/fed with the inoculation with a mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* for the highest CO_2 evolution in the rhizosphere under sunflower plants in both seasons irrespective of the non-significant effects of some interactions.

Table 4 deals also with the measurements of dehydrogenase activity (DHA) at sunflower flowering stage as affected by the applied treatments in 2016 and 2017. It can be comprehensively said that the effects of sole variables held the same as those observed with CO_2 evolution in both seasons of the current study. In other words, there existed the following significant descending order of AS> AN> urea-f with significantly better application rate of 60kg N/fed with inoculation with a mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* for the highest DHA in the rhizosphere under sunflower plants in both seasons irrespective of the non-significant effects of some interactions in both seasons of the current study. This combination treatment effected for the highest growth rate of microbes whether native in the soil and organic manure or being inoculated into the rhizosphere under sunflower plants in the new valley area.

TABLE 4. CO_2 evolution and dehydrogenase activity at sunflower flowering stage as affected by the applied treatments in 2016 and 2017

N forms	Bio	CO_2 evolution (mg/ 100gm dry soil/ 24hrs)				Dehydrogenase activity (DHA) (µg TPF/ g dry soil/ 24hrs)			
		2016		2017		2016		2017	
		60kg N	30kg N	60kg N	30kg N	60kg N	30kg N	60kg N	30kg N
AS	Control	3.83 j	3.47 k	5.27 k	4.62 m	7.20 j	6.64 l	7.62 j	6.78 n
	Azotobacter	5.50 e	5.14 f	7.23 e	6.80 f	8.67 d	8.58 d	9.17 c	8.65 e
	Azospirillum	5.03 f	4.70 g	6.73 f	6.20 hi	8.18 f	7.63 g	8.85 d	8.20 h
	Mixed	7.60 a	7.07 b	9.40 a	9.17 a	9.82 a	9.15 c	10.13 a	9.70 b
AN	Control	2.80 l	2.20 m	5.03 kl	4.30 n	6.35 m	6.17 n	6.92 m	6.37 o
	Azotobacter	4.93 fg	4.30 h	7.13 e	6.47 fg	8.27 f	7.70 g	8.67 e	7.83 i
	Azospirillum	4.27 h	3.73 j	6.53 fg	5.93 ij	7.67 g	7.38 h	8.35 g	7.75 i
	Mixed	6.18 c	5.80 d	8.73 b	8.50 b	9.28 b	8.70 d	9.62 b	9.10 c
Urea-F	Control	2.30 m	2.07 m	4.77 lm	3.87 o	5.60 o	5.10 p	6.35 o	5.85 p
	Azotobacter	4.17 hi	4.00 ij	6.43 gh	6.23 h	7.73 g	7.25 ij	7.78 i	7.17 l
	Azospirillum	3.85 j	3.40 k	5.80 j	5.23 k	7.37 hi	6.77 k	7.38 k	6.90 m
	Mixed	5.53 e	5.17 f	8.03 c	7.77 d	8.68 d	8.45 e	8.82 d	8.50 f

Mean values in the above table sharing one alphabet are not significantly different.

Plant height, number of leaves per plant, head diameter, and head dry weight as plant traits are presented in the following to support illustrating how the applied treatments could impose their influence on plant potential to respond to those treatments.

Plant height and number of leaves

Data in Table 5 show clear-cut effects on sunflower plant height that are as close as those exposed by the effects of applied treatments on soil available N, P, and K shown previously in table 2. Comprehensively, the best combination treatment was that composed of applying AS at the rate of 60kg N/fed and inoculating with a mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* to produce the highest plants of sunflower grown on the soil of the new valley area. This finding held true for the two seasons of the current study. In conclusion, superiority of this combination treatment to accumulate the highest available N, P, and K and promoting the highest microbial counts of both *Azotobacter* and *Azospirillum* as pointed to by both of highest CO_2 evolution and DHA in the rhizosphere under sunflower plants could acquire the highest potential for producing the highest plant height.

This finding held true for the measurements of number of leaves/plant in both seasons of study irrespective of the existence of some non-significant 2-variable interactions and the 3-variable interaction in one or both seasons of study.

Head diameter and head dry weight

Data in Table 6 show clear-cut significant effects on sunflower head diameter related to N forms and biological fertilizers in both seasons of study. The higher application rate could not show significant difference from the lower one on the measurements of sunflower head diameter in both seasons of study. This finding declares that applying N forms at 60kg N/fed was as effective as that at 30kg N/fed. Albeit economic in saving the amount of applied N sources, the previous plant traits discussed impose the ideology of applying them at the higher rate. Despite that some interactions could not show up as significant on the measurement of head diameter, the best combination treatment was that composed of applying AS at the rate of 60kg N/fed and inoculating with a mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* to produce the highest head diameter of sunflower grown on the soil of the new valley area in both seasons of study.

Data in Table 6 tells also that the sunflower head dry weight could exhibit significant effects due to the application of N sources, application rate, and biofertilizer variables. Comprehensively,

the best combination treatment could be figured out as applying AS at the rate of 60kg N/fed and inoculation with a mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* to obtain the highest dry weight of sunflower head.

Plant dry matter production, seed yield, and oil yield:

Data in Table 7 show clear-cut significant effects on sunflower dry matter production in both seasons of study. In other words AS significantly surpassed AN which, in turn, surpassed urea formaldehyde in producing the highest dry matter of sunflower plants. The higher rate significantly surpassed the lower in both seasons of study. With regard to bio-fertilization, both *Azotobacter chroococcum* and *Azospirillum lipoferum* surpassed the un-inoculated control treatment, with *Azotobacter* being the better of the two inoculants in effecting for the highest dry matter production of sunflower plants in both seasons. Amazing was that applying both inoculants together as mixed treatment surpassed the performance of both inoculants when they are separately applied. From another stand point, the three 2-variable interactions were consistently not significant. In other words these three interactions, could not translate the advantages that were described previously in this current research into significant influences on sunflower plant dry matter production in both seasons of study. This may be attributed to the clear-cut significant effects of sole variable of N sources, rates of application, and biological fertilizers on the production of dry matter production. It may also reflect the plants shifting to induce pollination and seed initiation and filling with redirection of plant manufactured food to perform seed maturation. Therefore, plants perform the senescence action through which they shift all nutrients from plant parts and move them toward seeds. This attitude seems to be emphasized by the observation regarding the 3-variable interaction which showed up to be significant in producing dry matter of sunflower plants. Comprehensively, the best combination treatment was, consistently, that composed of applying AS at 60kg N/fed and inoculation with bio-fertilizer mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* to produce the highest significant dry matter by sunflower plants.

Data in Table 7 tells also that seed yield showed, more or less, the same influences of N sources and biofertilizer variables showed up as significant as those with plant dry weight as mentioned above. While the effect of application rates could not translate itself into significant effect on seed production. This may be referred to that the activities of the applied biofertilizers compensated for the gap in performance between the two adopted rates, especially in the final stages of plant life span. In other words, the fixed atmospheric N by the biofertilizers could mimic or hinder or make up the difference between the two rates of application. Comprehensively, the best combination treatment was, consistently, that composed of applying AS at 60kg N/fed and inoculation with bio-fertilizer mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* to produce the highest significant seed weight per plant of sunflower.

With respect to oil yield of sunflower plants grown on the soil of the new valley area, Table 7 tells also that oil yield reflected the directions of all applied treatments and their interactions, except for one interaction which is that between the application rate and the bio-fertilizers, which was not significant, in both seasons of study. Generally, the best combination treatment was, consistently, that composed of applying AS at 60kg N/fed and inoculation with bio-fertilizer mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum* to produce the highest oil yield of sunflower plants.

Seed N, P, and K contents of sunflower

The calculated effects of single variables; N forms, rates of application, and bio-fertilizers, could be significant on accumulating N, P, and K in seeds of sunflower plants in both seasons of study as shown in Table 8.

In other words, AS was significantly better than AN, which, in turn, was better than urea-formaldehyde in inducing the highest concentrations of the three nutrients in sunflower seeds. Being the lowest in salt index but highest in acidifying the rhizosphere, AS could also render mild salt effect on the roots of the grown plants with proliferation of more available plant nutrients. In addition, the higher rate of N forms application was significantly better than the lower one. Inoculation with free-living microbes could consistently prove being significant on seed content of the three nutrients as compared with the control treatment. *Azotobaacter chroococcum* was significantly better than *Azospirillum lipoferum* in

this respect when they are applied separately, but, the mixed application of both microbes together greatly surpassed applying them separately. The three 2-variable interactions could not play consistent role in accumulating the three nutrients in sunflower seeds. They showed intermittent significant pattern. The 3-variable interaction showed consistent non-significant role in this regard. Generally, the best combination treatment was, consistently, that composed of applying AS at 60kg N/fed and inoculation with bio-fertilizer mixture of *Azotobacter* and *Azospirillum* to accumulate the highest N, P, and K nutrients in the seeds sunflower plants.

TABLE 5. Plant height (cm) and number of leaves/plant prior to sunflower harvest as affected by the applied treatments in 2016 and 2017

N forms	Bio	Plant height (cm)				No. of leaves/plant			
		2016		2017		2016		2017	
		60kg N	30kg N	60kg N	30kg N	60kg N	30kg N	60kg N	30kg N
AS	Control	154 g	139 j	155 j	143 l	13 k	9 mn	17 kl	14 n
	Azotobacter	193 b	168 e	184 b	177 f	30 b	25 e	32 bc	27 f
	Azospirillum	171 d	161 f	185 d	168 g	27 cd	23 f	29 e	25 g
	Mixed	197 a	178 c	202 a	187 c	34 a	30 b	37 a	31 cd
AN	Control	97 p	83 q	106 r	91 s	10 lm	8 n	16 lm	11 hi
	Azotobacter	161 f	146 h	165 h	149 k	25 e	21 gh	30 de	22 i
	Azospirillum	153 g	143 i	162 i	143 l	22 fg	20 h	25 g	27 f
	Mixed	171 d	161 f	183 e	166 h	28 c	26 de	33 b	27 f
Urea-F	Control	62 r	52 s	66 t	56 u	10 lm	8 n	10 o	8 p
	Azotobacter	117 l	106 n	137 m	121 p	18 i	16 j	20 j	15 mn
	Azospirillum	113 m	102 o	128 o	119 q	16 j	14 k	18 k	15 mn
	Mixed	`142 i	125 k	148 k	133 n	23 f	20 h	24 gh	22 i

Mean values in the above table sharing one alphabet are not significantly different.

TABLE 6. Head diameter (cm) and dry weight (gm) prior to sunflower harvest as affected by the applied treatments in 2016 and 2017

N forms	Bio.	Head diameter (cm)				Head dry weight (gm)			
		2016		2017		2016		2017	
		60kg N	30kg N	60kg N	30kg N	60kg N	30kg N	60kg N	30kg N
AS	Control	21 g	18 i	23 j	19 n	9 f	6 h	9 h	8 i
	Azotobacter	31 b	23 e	33 b	27 f	13 b	11 d	15 b	12 e
	Azospirillum	30 c	11 o	32 c	25 h	12 c	10 e	14 c	11 f
	Mixed	34 a	18 i	36 a	31 d	14 a	14 a	16 a	15 b
AN	Control	15 l	13 m	16 p	16 p	7 g	6 h	9 h	7 j
	Azotobacter	19 h	18 i	24 i	24 i	12 c	10 e	13 d	11 f
	Azospirillum	18 i	17 j	21 l	21 l	11 d	9 f	13 d	11 f
	Mixed	23 e	22 f	28 e	26 g	14 a	13 b	14 c	13 d
Urea-F	Control	11 o	12 n	13 r	14 q	6 h	5 i	7 j	6 k
	Azotobacter	18 i	16 k	20 m	18 o	11d	9 f	13 d	10 g
	Azospirillum	15 l	15 l	18 o	16 p	11 d	9 f	12 e	10 g
	Mixed	19 h	18 i	23 j	22 k	12 c	12 c	14 c	13 d

Mean values in the above table sharing one alphabet are not significantly different.

TABLE 7. Plant dry weight (gm), seed yield (kg/fed), and oil yield (kg/fed) of sunflower as affected by the applied treatments in 2016 and 2017

N forms	Bio	Plant dry weight (gm)				Seed yield (kg/fed)				Oil yield (kg/fed)			
		2016		2017		2016		2017		2016		2017	
		60	30	60	30	60	30	60	30	60	30	60	30
AS	Control	369 de	321 gh	381 ef	330 h	82 hi	78 hij	87 k	80 lm	369 de	321 gh	381 ef	330 h
	Azotobacter	393 b	367 de	411 bc	393 de	125 b	114 de	135 c	123 e	393 b	367 de	411 bc	393 de
	Azospirillum	385 c	361 e	391 de	373 f	121 bc	103 f	128 d	114 fg	385 c	361 e	391 de	373 f
	Mixed	413 a	414 a	424 a	418 ab	139 a	123 bc	155 a	140 b	413 a	414 a	424 a	418 ab
AN	Control	280 kl	265 m	289 k	260 n	67 k	67 k	78 mn	75 n	280 kl	265 m	289 k	260 n
	Azotobacter	336 f	293 j	350 g	309 ij	106 ef	84 gh	116 f	108 h	336 f	293 j	350 g	309 ij
	Azospirillum	331 fg	289 jk	345 g	302 j	89 g	80 hi	110 gh	100 i	331 fg	289 jk	345 g	302 j
	Mixed	375 cd	313 hi	402 cd	373 f	117 cd	107 ef	126 de	122 e	375 cd	313 hi	402 cd	373 f
Urea-F	Control	242 o	242 o	240 o	245 o	72 jk	66 k	68 o	64 o	242 o	242 o	240 o	245 o
	Azotobacter	277 l	264 mn	281 kl	277 klm	85 gh	77 ij	92 j	84 kl	277 l	264 mn	281 kl	277 klm
	Azospirillum	270 lm	253 no	270 lmn	267 mn	78 hij	70 k	88 jk	79 mn	270 lm	253 no	270 lmn	267 mn
	Mixed	308 i	295 j	348 g	319 hi	105 f	89 g	117 f	100 ij	308 i	295 j	348 g	319 hi

Mean values in the above table sharing one alphabet are not significantly different.

TABLE 8. N, P, and K contents of sunflower seeds as affected by the applied treatments in 2016 and 2017

N forms	Bio.	Seed N (µg N/gm)				S P (µg P/gm)				S K (µg K/gm)			
		2016		2017		2016		2017		2016		2017	
		60	30	60	30	60	30	60	30	60	30	60	30
AS	Control	2.82 i	2.40 kl	2.83 m	2.33 n	0.29 j	0.23 o	0.33 m	0.27 r	0.71 m	0.62 e	0.74 l	0.67 n
	Azotobacter	3.57 bc	3.37 de	3.68 bcde	3.48 fg	0.40 c	0.33 h	0.44 d	0.38 h	0.87 c	0.83 e	0.92 b	0.85 f
	Azospirillum	3.28 ef	3.03 g	3.32 h	3.13 ij	0.55 g	0.31 i	0.4 e	0.34 l	0.81 f	0.77 i	0.86 e	0.80 i
	Mixed	3.95 a	3.57 bc	4.07 a	3.70 bcd	0.43 a	0.36 f	0.53 a	0.46 c	0 92 a	0.88 b	0.95 a	0.89 c
AN	Control	2.57 j	2.33 l	3.03 jkl	2.28 n	0.25 m	0.20 q	0.29 p	0.24 s	0.65 n	0.57 q	0.71 m	0.65 o
	Azotobacter	3.40 de	3.13 fg	3.57 def	3.35 gh	0.37 e	0.31 i	0.41 f	0.37 i	0.84 d	0.80 g	0.86 e	0.82 g
	Azospirillum	3.10 g	2.87 hi	3.15 ij	3.07 jk	0.33 h	0.27 k	0.40 g	0.32 n	0.77 i	0.73 l	0.81 h	0.78 j
	Mixed	3.63 b	3.45 cd	3.82 b	3.63 cde	0.41 b	0.35 g	0.48 b	0.44 d	0.88 b	0.84 d	0.92 b	0.86 e
Urea-F	Control	2.52 jk	2.28 l	2.97 kl	2.22 n	0.24 n	0.21 p	0.28 q	0.22 t	0.63 o	0.56 r	0.65 o	0.62 p
	Azotobacter	3.33 de	3.10 g	3.48 fg	3.22 hi	0.36 f	0.29 j	0.38 h	0.35 k	0.79 h	0.76 j	0.82 g	0.80 i
	Azospirillum	3.02 gh	2.75 i	3.07 jk	2.90 lm	0.31 i	0.26 l	0.36 j	0.31 o	0.74 k	0.71 m	0.78 j	0.77 k
	Mixed	3.58 bc	3.37 de	3.75 bc	3.55 def	0.39d	0.33 h	0.46 c	0.41 f	0.87 c	0.83 e	0.88 d	0.85 f

Mean values in the above table sharing one alphabet are not significantly different.

Conclusion

This research work was conducted in two successive seasons (2016/2017) at the new valley research station of the Desert Research Centre (DRC) on a soil characterized by hot atmospheric conditions. The aim of this work was to feed sunflower plants to attain good vegetative growth, achieve the highest possible seed and oil yields, and be able to combat or loop around the adverse effects dominating the study area. The current study found that the combination treatment composed of applying AS at 60kg N/fed and inoculating with bio-fertilizer mixture of *Azotobacter chroococcum* and *Azospirillum lipoferum*, which is capable of fixing atmospheric nitrogen, then release it into the rhizosphere hosting plant roots, could proliferate highest soil available N, P, and K in the rhizosphere under sunflower plants. It could be significantly accommodate the highest biological activity that was evidenced by highest total *Azotobacter chroococcum* counts, highest CO_2 evolution, and highest dehydrogenase (DHA). This treatment could be prelude to the highest plants with highest number of leaves per plant, largest head diameter and head dry weight, plant dry matter production, seed yield, and oil yield. Same treatment could accumulate N, P, and K in sunflower seeds. Apparently, this combination treatment seemed able to combat the adverse environmental conditions dominating the study area at the new valley region.

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تأثير اللقاحات الحيوية والصور المختلفة من الأسمدة النيتروجينية على إنتاجية وجودة نبات دوار الشمس تحت ظروف الوادى الجديد

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تم تنفيذ البحث خلال موسمين متتاليين عامى 2016/2017 بمحطة بحوث الوادي الجديد التابعة لمركز بحوث الصحراء. كان الغرض من إقامة هذا البحث هو محاولة تغذية نباتات دوار الشمس للحصول على نمو جيد وإنتاجية مرتفعة من المحصول، وذلك من خلال تأثير الأنشطة الميكروبية للمصادر التى تم إضافتها فى التربة (*Azotobacter chroococcum* and *Azospirillum lipoferum*) سواء منفردة أو عند تفاعلها مع مصادر الأسمدة الأزوتية المستخدمة (نترات الأمنيوم، سلفات الأمونيوم، يوريا فورمالدهيد).

ولقد توصلت الدراسة إلى أن المعاملة السمادية المركبة من إضافة سماد سلفات الأمونيوم بمعدل 60 كجم/فدان مع التلقيح بخليط من ميكروبي الآزوتوباكتر والآزوسبيريلليم ذات المعيشة اللاتكافلية في منطقة انتشار الجذور والتى تقوم بتثبيت النتروجين الجوي في منطقة الريزوسفير المحيطة بجذور النباتات هى الأفضل في تثبيت الآزوت الجوى بسبب الزيادة المؤكدة معنوياً في أعداد هذه الميكروبات مما أدى إلى زيادة ذوبان كل من الفوسفور والبوتاسيوم. وقد دل على ذلك أيضا زيادة انطلاق ثاني أكسيد الكربون وكذلك زيادة نشاط انزيم الديهيدروجينيز في منطقة انتشار الجذور بدرجة مؤكدة معنوياً بالمقارنة بالمعاملات الأخرى. وقد مهدت هذه المعاملة أيضاً للوصول إلى أعلى ارتفاع للنباتات و أكبر عدد من الأوراق على النبات، وأكبر قطرٌ ووزن للقرص، وأعلى إنتاجيةً للمادة الجافة، وأعلى محصولاً للبذور، وكذلك أعلى محصولاً للزيت الناتج، كما وقد تمكنت هذه المعاملة المركبة من إحداث أعلى تجمع لبعض العناصر مثل النيتروجين، والفوسفور، والبوتاسيوم في البذور الناتجة. ويبدو ظاهرياً أن هذه المعاملة قد مكنت النباتات من النمو الجيد والمحصول المرتفع وأيضا التغلب على الظروف الصعبة السائدة بمنطقة الدراسة بالمقارنة بالمعاملات الأخرى في هذا العمل البحثي.