Effect Of Biofertilization By Using Three Azotobacter Isolates And Two Levels Of Mineral Nitrogen Fertilizer On Jerusalem Artichoke (*Helianthus Tuberosus* L.,) Growth, Yield And Some Chemical Constituents.

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ABSTRACT: The present investigation was carried out at the laboratory of Biofertilizer Center, Minia University and the Experimental Farm, Fac. of Agric., Minia University during 2011 and 2012 seasons to study the effect of N fertilizer, three Azotobacter isolates and their interactions on growth yield and chemical composition of Jerusalem artichoke (*Helianthus tuberosus* L.,).The use of Isolate 3+50% recommended dose of **N.** fertilization gave the highest level of shoot fresh weight in both seasons. The results also showed that use isolate 3 was more efficient than neither Isolate 1 nor Isolate 2 for increasing fresh weight. The same trend was also observed when shoot dry weight was studied. Data of plant height (cm) indicates that use of Isolate 3+50% chem gave significant effect on plant height in both seasons . The reported results showed that the bacterial isolates improved plant height and some other vegetative characters of Jerusalem artichoke plants only when 50% of the N recommended dose were applied. These results showed that the values of number of branches/plant in all treatments don't exceed those recorded for control. The highest values of tubers weight on fresh basis were recorded when Isolate 3+50% N was applied (4.9 kg fresh weight basis (fwb)) in the first season and (4.83 kg fwb) in the second season whereas the lowest ones was recorded for control +50% chem. (. 3.2 kg fwb) The heaviest tuber fresh weight values were 86.13 and 91.43 g for treatment included Isolate 2+50% chem. in the first and second seasons, respectively. The treatments applied in the present work led to significant increases in the weight of single tuber with different extents ranged from 208% to 264%. The results of the dry weight of 100 g of tuber showed that all treatments increased this character except control+50% N which decreased this character (22.43g). The effect of the bacterial isolates plus N fertilization at 50% dose ordered as follows: Isolate 1 > Isolate 2 > Isolate 3 whereas at 100% N fertilization where Isolate 2> Isolate 1> Isolate 3. Accumulation rate of dry matter was highest in tubers treated with Isolate 1+100% chemical fertilizer followed by treatment-3 Isolate 2 + 100% chemical fertilizer). The results showed that combining Azotobacter isolates and chemical N fertilization increased dry matter by 181% when compared with control. The concentrations of total phenolic compounds (TPCs) in whole tuber extracts of Treatment-4 which formed from Isolate 1+100% N were higher (52.5 mg/100g fw) than those determined in control with extent reached to be 235%. The concentrations of total flavonoids (TFs) ranged from 56 to 145 µg/100 g fwb in control sample and Treatment-4 respectively. The results also, showed that the differences between treatment-3, treatment-4 and treatment-6 are not sharp and ranged from 139 to 145 µg/100 g fwb. The total soluble sugars (TSS) assayed in the samples ranged from 6.9 to 11.6 % in untreated sample and Treatment-6 respectively.
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Key words : Jerusalem artichoke, *Helianthus tuberoses*, Biofertilization*, Azotobacter,* Phenolic compound, Falvonids, Isolates.

1-INTRODUCTION:

Jerusalem artichoke, (*Helianthus tuberosus* L.) is an agricultural crop with a great potential for high sugar yield per hectare (9-13 t/ha,) Klaushofer 1986) and could be grown in all planting dates, (Puangbut, *et al*.,2012). Jerusalem artichoke contains an important homopolysaccharide (Inulin) which play very important roles for nutrition and treatment of diabetic patients and obesity. Recently, Jerusalem artichoke has attracted attention by the bioethanol industry sector because of its high productivity as well as high content of inulin (Puangbut, *et al*., 2012). The inulin is present as a reserve carbohydrate in its tubers and can be easily hydrolyzed for ethanol production (Toyohiko *et al*., 1996; Mario *et al*., 2004; Maria *et al*., 2006).Using Biofertilizers has increased with increasing the agricultural production.

Handling of Jerusalem artichoke plant tubers are similar to that of potatoes. Planting cut pieces of the tubers contains one or two eyes under optimum conditions, may produce equal or nearly equal that obtained from whole tubers (Klug–Anderson (1992). Meanwhile, there is a risk that serious reductions in yields may be caused by mistakes in the cutting, storage and planting of the seed-pieces, However, climatic conditions unfovaurable to the healing of cut

tubers, could be increased the incidence of diseases which attack the cut surfaces (Bolye and Baukwill, 1955; El-Sharkawy, 1998).

Biological nitrogen fixation plays positive and constructive role in maintaining soil N. In the past green revolution, intensive monocropping, excessive N-fertilizer use and maximum output with minimum input has deteriorated soil health. So it needs judicious use of chemical fertilizers in combination with the use of efficient, effective and competitive Rhizobium and other nitrogen fixing inoculants (Dobrei *et al*., 2001).

Biofertilization will help bring down the costs of chemical fertilizers especially N and P and improves soil fertility by maintaining the physical conditions of the soil. Biofertilizers consist mainly of beneficial microorganisms that can release nutrients from raw materials and plant residues in the soil and make them available, commercially and specific strains such as *Azotobacter* isolates and *Azospirillum sp*. are used as biological fertilizer (Pathak *et al*., 1997 and Ribaudo *et al.*, 2006).

In spite of the huge additions of chemical N fertilizers to the cultivated soil in Egypt, the available N level for plants is usually low, since it is may be lost from soil by volatilization, leaching, erosion and denitrification (Sathya et al., 2009). Biofertilizers, recently became a positive alternative to chemical fertilizers. They are safe for human, animal and environment and using them was accompanied with reducing the pollution occurred in our environment. They may help in improving crop productivity and quality by increasing the biological N fixation, the availability and uptake of nutrients and stimulating the natural hormones (El-Haddad *et al*., 1993 and Subba-Rao *et al*., 1993). Application of biofertilizers achieved the following merits: (1) reducing plant requirements of nitrogen by 25%, (2) improving the availability of various nutrients for plant absorption, (3) increasing the resistance of plants to root diseases and (4) reducing the environmental pollution by (Marangoni et al ., 2001, Foly et al .,2002 , and Kannaiyan ,2002) .

Phenolic compounds are commonly found in both edible and non-edible plants, and they have been reported to have multiple biological effects. Crude extracts of vegetables such as Jerusalem artichoke**,** potato and tomato rich in phenolics are increasingly of interest in the food industry because they retard oxidative degradation of lipids and thereby improve the quality and nutritional value of food (Kahkönen *et al*., 1999).

In general, plants contain a certain degree of resistance against insect predation, which is reflected in the limited number of insects capable of attacking this plant. This resistance is the result of a set of defence mechanisms acquired by plants during evolution (Schuler *et al*., 1998). In general, many secondary chemical compounds have been associated with plant defense. (Hilder *et al*., 1987).

All members of the family Azotobacteraceace can fix, atmospheric nitrogen, Azotobacters are aerobic, mainly soil-dwelling organisms with allniquc array of metabolic capabilities in addition to nitrogen fixation. Different species and strains have various abilities to synthesize alginates, polyhydroxybutyrate (P-$_{OH}$B) and plant hormones (Safwat *et al*., 2008). Thus, the aim of the present investigation is to study the effect of N fertilizer, some Azotobacter isolates and their interactions on growth characters, yield and yield components and chemical composition of Jerusalem.

2.MATERIAL AND METHODS

The present investigation was carried out at the laboratory of Biofertilizer Center, Minia University and the Experimental Farm, Fac. of Agric., Minia University during 2011 and 2012 growing seasons to study the effect of two levels of N fertilizer, three Azotobacter isolates and their interactions on growth and chemical composition of Jerusalem.

2.1.Materials:-

2.1.1. Isolates

Azotobacter three isolates of Azotobacter (1, 2 and 3) were kindly obtained from the Biofertilizer Center, Minia University .

2.1.2. Media:-

Complete media (Standberg and Wilson 1968) was used for growing Azotobacter.

2.1.3 Mineral N Fertilizer :

Nitrogen levels (50 % and 100% of the recommended dose (50 Kg / feddan were provided and ammonium nitrate (20.6 %N) was applied in two equal doses N) after 30 and 60 days from planting.

2.2.Methods:

2.2.1. Preparation of inoculation:-

Three flasks, each containing 500 ml liquid complete medium, were inoculated with one isolate and then incubated at 30°C for 4 days. The condensed growth in each flask (10^8 cell/ml) was diluted in 5.0 L irrigated river water. Inoculation of *Azotobacter* cells in each experimental plate was performed by Biofertilizer Center , Minia University , dripping the diluted suspension twice ; after 15 and 20 days of seed sowing.

Randomized Complete Block Design (RCBD) with three replications was used in both seasons.

The seed tubers of Jerusalem artichoke (*Helianthus tuberosus* L.,) cv. Local were kindly obtained from the Institute of Horticulture, Center of Agriculture, Giza, Egypt. The tubers were sown on 15 and 18 April in the first and second season, respectively.

Three isolates of Azotobacter were used as solution and added after 15 and 20 days from tuber sowing in

three holes around each plant, in addition to the control that was irrigated water. The 8 tested treatments were:

1- Azotobacter isolate 1 + 50% of the recommended nitrogen dose
2- Azotobacter isolate 1 + 100% of the recommended nitrogen dose
3- Azotobacter isolate 2 + 50% of the recommended nitrogen dose
4- Azotobacter isolate 2 + 100% of the recommended nitrogen dose
5- Azotobacter isolate 3 + 50% of the recommended nitrogen dose
6- Azotobacter isolate 3 + 100% of the recommended nitrogen dose
7- Irrigated water + 50% of the recommended nitrogen dose
8- Irrigated water + 100% of the recommended nitrogen dose

The plants were also received calcium superphosphate (15.5% P_2O_5) and potassium sulphate (K2SO4) at the recommended dose of potato commercial production. The other agricultural practices were done as in commercial growing potato crop.

2.2.2.Record of Growth and yield parameters :

Data were recorded for the following parameters**:**

1. Shoot fresh weight, g
2. Shoot dry weight, g
3. Plant height,cm
4. No. of branches/plant
5. Tuber fresh weight/plant
6. Average tuber weight, g
7. Dry weight of 100 gm of fresh tuber, g

At harvesting, the chemical constituents of all combined treatments from treatment 1 to treatment 6 as well the control which represented by 100% of the recommended nitrogen dose were determined as follows:

A-Chemical composition:-

Approximate chemical analyses, moisture, dry matter and total ash content of Jerusalem artichoke samples were carried out according to Official Methods of the Association of Official Analytical Chemists (AOAC, 1975). All determinations were performed in triplicates and their means were reported.

B-Extraction and determination of (TPCs):-

The total phenolic compounds (TPCs) were extracted from each fresh sample (1.0g) by the method described by Taga *et al*., (1984) The amount of TPCs is estimated as tannic acid equivalent according to the Folin-Ciocalteu method as described by Singleton and Rossi (1965).

C-Extraction of total flavonoids (TFs):-

Fresh Jerusalem artichoke samples (3.0 g) were extracted in a Soxhlet extractor with 100 ml distilled water or ethanol for 1 hour and the extract filtered. TFs was measured spectrophotometerically by the method described by Zhuang *et al*., 1992).

D-Extraction and determination of TSS:-

Total Soluble Sugars (TSS) extracted from fresh tubers of Jerusalem artichoke (2.0g) were done according to Macrae and Zand-Moghdlam (1978) method. TSS were determined by the phenol-sulfuric acid method as described by Dubois *et al*, (1956).

2.2.4.Statistical analysis:-

Data were subjected to analysis of variance procedures and means were compared using L.S.D. test according to Gomes and Gomes, (1984).

3.RESULTS

3.1. Effect of use three Azotobacter isolates and chemical fertilization on shoot fresh weight:

Results recorded in Fig. (1) and Table (1) showed that the use of Isolate 3 + 50% chem. N gave the highest level of shoot fresh weight (kg) and the lowest one was for Isolate 3 + 100% N . in both seasons. The results also showed that use isolate 3 was more efficient than Isolate 1 and Isolate 2 for increasing fresh weight . Thus, variability was very high among the tested isolates for this trait .

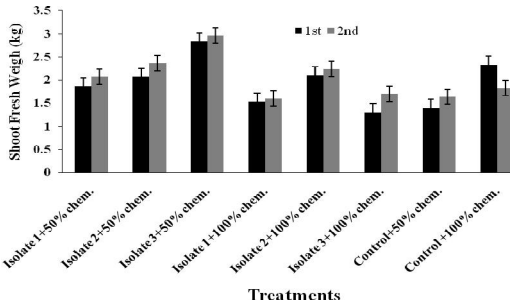

Fig. (1): Effect of use three Azotobacter isolates and chemical fertilization on shoot fresh weight

Reust and Dutoit (1992) found in the *Helianthus tuberosus* L trials that a basal fertilizer application of 80 Kg P_2O_5 + 240 Kg K_2O/ha supplemented by up to 40 Kg N mineral or organic fertilizer at planting, produced tuber yields which ranged from 25.0 to 47.52 t/ha, DM content from 19 to 24%, tuber sugar contents from 11 to 19% and shoot and leaf fresh weight ranged from 5.0 to 48.0 t/ha. Also, results recorded by Gao, *et al*., (2011) showed significant positive correlation between leaf and root size and insignificant correlation between leaf and tuber size.

3.2 Effect of use three Azotobacter isolates and chemical fertilization on shoot dry weight

Data of this character are shown in Fig. (2) and Table (1). It is clear from these results that Isolate 3+50% chem. treatment produced the highest shoot dry weight.

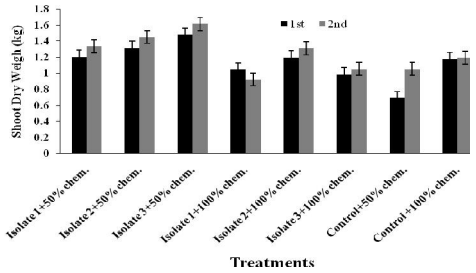

Fig. (2): Effect of use three Azotobacter isolates and chemical fertilization on shoot dry weight

Table (1): Effect of three isolates and two levels of chemical fertilization on four different vegetative characters of Jerusalem artichoke

Treatments	Shoot fresh weight (kg)		Shoot dry weight (kg)		Plant height (cm)		No. of branches/plant	
	1st	2nd	1st	2nd	1st	2nd	1st	2nd
Isolate 1+50% chem.	1.867BCD	2.067 BC	1.203BC	1.335BC	176.0 DE	185.7C	26.00C	30.67 DE
Isolate 2+50% chem.	2.067BC	2.367 B	1.317 AB	1.452B	188.0 BCD	181.3C	30.67BC	36.67BC
Isolate 3+50% chem.	2.833A	2.967 A	1.478A	1.613A	204.7A	218.7 A	34.00B	38.67AB
Isolate 1+100% chem.	2.100BC	2.233 B	1.198 ABC	1.312CD	192.7ABC	202.3B	22.00D	28.33E
Isolate 2+100% chem.	1.533CD	1.600 D	1.047BC	0.9217 E	169.3E	175.7 C	22.00D	34.67BCD
Isolate 3+100% chem.	1.300D	1.700D	0.9883 C	1.055E	183.3CDE	179.3C	29.67BC	34.33 CD
Control+50% chem.	1.400CD	1.633 D	0.6900 D	1.055 E	184.0CDE	182.3C	35.33B	34.33CD
Control +100% chem.	2.333AB	1.833CD	1.176 BC	1.195 D	201.0 AB	204.3B	42.67A	41.67 A
LSD 0.05	0.7135	0.3181	0.2878	0.1356	14.95	11.54	6.004	4.006

3.3 Effect of use three Azotobacter isolates and chemical fertilization on plant height (cm):

Data of plant height (cm) shown in Fig. (3) and Table (1). The results indicate that use of Isolate 3+50% chem gave significant effect on plant height both seasons. The use of Isolate 3+100% chemical fertilization gave the lowest level of plant height. El-Sharkawy *et al*., (1998) studied the effect of cultivar on stem length and they found that Fuseau cultivar gave higher stem length than the local cultivar and they added also that planting whole tubers produced higher stem length than that
produced from cut tubers in two successive seasons.

Results showed that the bacterial isolates in this work improved plant height and some other vegetative characters of Jerusalem artichoke plants only when only 50% of the chemical N fertilization was applied . Gao, *et al*., (2011) studied the response of plant height of Jerusalem artichoke (*Helianthus tuberosus* L.) to water and nitrogen fertilization treatments in China. They found that water was a main limiting factor on the height and yield of Jerusalem artichoke. It significantly improved the height and yield of Jerusalem artichoke (including leaf, stem, root and tuber yields) ($p<0.01$). Also, they concluded that nitrogen fertilizer significantly influence plant height and yield.

Klug-Andersen (1992), reported that in Jerusalem artichoke, different planting materiel sizes (25-200 g/tubers) did not affect the tuber characters investigated. Jerusalem artichoke (*Helianthus tuberous* L., cv. Topianka and cv. Violet de Rennes) were exposed to different supplies of N, P and K. Nitrogen supply increased tuber yield more than the weight of aerial parts. Nutrient regimes without P or K addition but including N were depressed the yield of tubers by 8-23% (Soja *et al*, 1994).

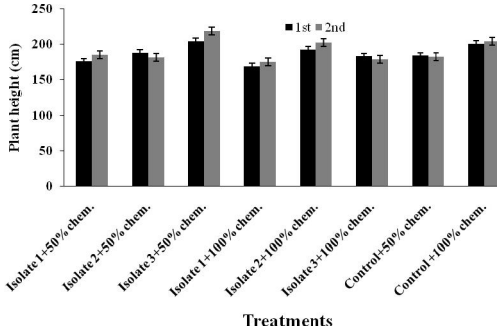

Fig. (3): Effect of use three Azotobacter isolates and chemical fertilization on plant height

3-4 Effect of use three Azotobacter isolates and chemical fertilization on number of branches/plant:

Data given in Fig. (4) and Table (1) generally showed that the number of branches/plant in the second season are higher than those reported in the first one except for control treatments. The obtained results also, showed that the values number of branches/plant in all treatments don't exceed those recorded for control.

El-Sharkawy *et al*., (1998) reported that whole tubers planting tubers gave higher No. of branches/plant than that of cut-tubers in both seasons. Concerning the effect of phosphorus levels and micro-elements on No. of branches, revealed that increasing the phosphorus level enhance number of branches/plant. Moreover, No. of branches were raised in each P level by spraying plants with the used micro-elements.

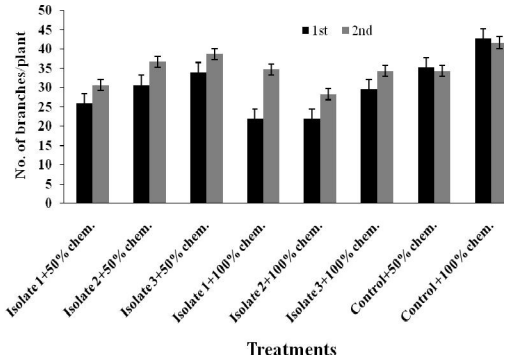

Fig. (4): Effect of use three Azotobacter isolates and chemical fertilization on No. of branches/plant.

3.5 Effect of use three Azotobacter isolates and chemical fertilization on tubers fresh weight/plant:

The highest values of tubers weight on fresh basis were recorded when Isolate 3+50% chemical fertilization was applied This treatment gave 4.9 kg fresh weight in the first season and 4.83 kg in the second season whereas the lowest ones were recorded for Control+50% chem. Which gave 3.2 in the first and 3.47 kg in the second season (Fig. 5+ Table 2). The obtained results showed that using this treatment increased the tuber weight by 150% and 139% with compared to control. The effect of water and nitrogen on weight of tubers of Jerusalem artichoke was studied by Gao, *et al*., (2011) and they found that tuber size and tuber weight were improved by nitrogen addition.

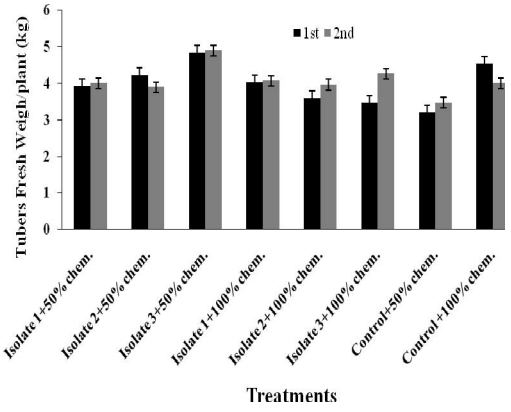

Fig. (5): Effect of use three Azotobacter isolates and chemical fertilization on Tubers fresh weight/plant

Table (2): Effect of three isolates and two levels of chemical fertilization on three different yield characters of Jerusalem artichoke

Treatments	Tuber fresh weight/plant, kg		Average tuber fresh weight (g)		Dry weight of 100 g of tubers fresh weight (g)	
	1st	2nd	1st	2nd	1st	2nd
Isolate 1+50% chem.	3.933 AB	4.000 B	77.20 A	77.30 C	31.77 A	31.47 A
Isolate 2+50% chem.	4.233 AB	3.900 B	86.13 A	91.43 A	30.73 AB	31.20 A
Isolate 3+50% chem.	4.833 A	4.900 A	72.57 A	82.70 B	26.20 C	27.20 BC
Isolate 1+100% chem.	4.033 AB	4.067 B	53.73 B	57.97 D	27.97 ABC	28.73 B
Isolate 2+100% chem.	3.600 AB	3.967 B	45.53 BC	47.57 EF	31.33 AB	30.60 A
Isolate 3+100% chem.	3.467 AB	4.267 B	33.83 C	41.70 G	25.70 C	27.30 BC
Control+50% chem.	3.200 B	3.467 C	43.90 BC	51.87 E	27.47 BC	22.43 D
Control +100% chem.	4.533 AB	4.000 B	32.57 C	43.97 FG	27.40 BC	26.87 C
LSD 0.05	**1.589**	**0.3837**	**14.39**	**5.383**	**3.949**	**1.675**

3.6 Effect of use three Azotobacter isolates and chemical fertilization on average tuber fresh weight (g):

The recorded results in Fig. 6 and Table 2 showed that the highest values of average tuber fresh weight are 86.13 and 91.43 g for Isolate 2+50% chem. N treatment in the first season and second one, respectively. These given results also showed that the values are always higher in second season than those found in the first one. The tested treatments applied in the present work led to great increases in the weight of tuber with different extents ranged from 208% to 264%

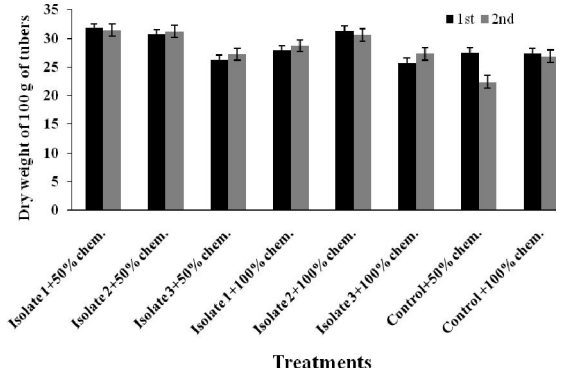

Fig. (7): Effect of use three Azotobacter isolates and chemical fertilization on dry weight of 100 g of fresh tuber

Klug-Andersen (1992), reported that in Jerusalem artichoke, different planting materiel sizes (25-200 g/tubers) did not affect the tuber characters investigated. Jerusalem artichoke (*Helianthus tuberous* L., cv. Topianka and cv. Violet de Rennes) were fertilized with different doses of N, P and K. They found that nitrogen supply increased tuber yield more than the productivity of aerial parts. Nutrient regimes without P or K addition but including N depressed the yield of tubers by 8-23% (Soja *et al*, 1994).

3.8 Effect of use three Azotobacter isolates and chemical fertilization on dry matter levels in *Jerusalem artichoke* plants.

The results of dry mater of whole tubers produced from all treatments as well as untreated sample are presented in Fig. (8). The results showed that the accumulation rate of dry matter was highest in tubers of treatment-4 which included Isolate 1+100% chemical fertilizer followed by treatment-3 which consists of Isolate 2+100% chemical fertilizer (25.4%). Our results also showed that use combining Azotobacter isolates and chemical fertilization led to increase of dry matter by 181% when compared to control. These results are in good agreement with those reported by El-Dahtory *et al*., (1989) and El-Sharkawy, 1998

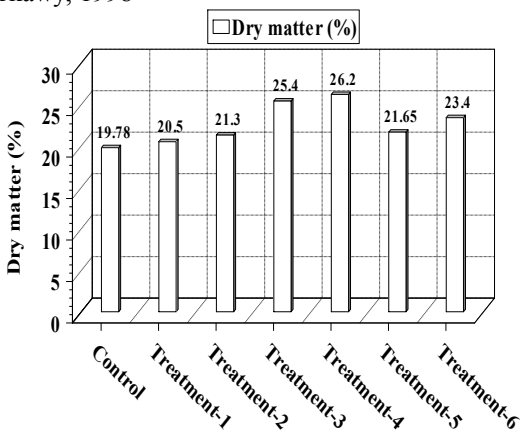

Fig. (8): Effect of use three Azotobacter isolates and chemical fertilization on dry matter levels in *Jerusalem artichoke* plants.

Results from fertilizer experiments frequently are inconsistent as the preexisting soil fertility plays an important role in the apparent nutrient requirement. Dorrell and Chubey (1977) found moderate or no yield increases due to increasing nutrient supply. In spite of the huge additions of chemical fertilizers to the cultivated soil in Egypt, the available nutrients level for plants is usually low, since it is rapidly converted to an unavailable form by its reaction with other soil constituents and conditions and becomes inaccessible by plants (El-Dahtory *et al*., 1989). Nitrogen is an essential element for plant growth and development.

3.9 Effect of use three Azotobacter isolates and chemical fertilization on TPCs:

The concentrations of total phenolic compounds (TPCs) in whole tuber extracts of Treatment-4 (Fig. 9) which formed from Isolate 1 + 100% chemical fertilizer were higher (52.5 mg/100g fw) than those determined in control with extent reached to be 235%. There are many reports concerning the functions of phenolic compounds in tuber plants such as potato, sweet potatoes and artichoke, these compounds play very important roles in defence mechanisms against pathogens and insects. The plants containing higher contents of TPCs are much resistant than those having lower concentrations (Franco *et al*., (2002). TPCs synthesize through secondary metabolism cycle and it could be produce some plant hormones. It is well known that bacteria of the genus *Azospirillum* synthesize auxins,

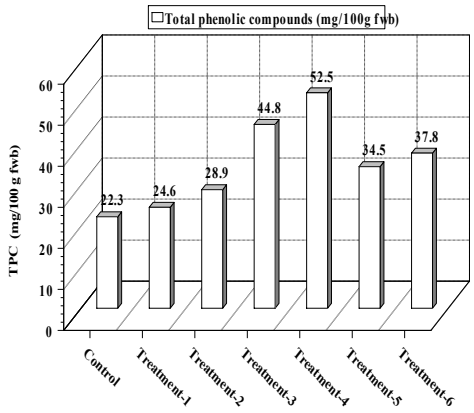

Fig. (9): Effect of biofertilization using three different isolates on total phenolic compound levels in Jerusalem artichoke plants.

especially indole-3-acetic acid (IAA) (Crozier *et al*., 1988), and variety of other auxins like indole-3-pyruvic acid, indole-3-butyric acid, and so on (Crozier *et al*., 1988; Costacurta *et al*., 1994). These bacterial compounds contribute to the plant auxin "pool" in such a way that the effect of *Azospirillum* inoculation can be minimized by exogenous auxin application (Glick *et al*., 1999). Production of auxins by azospirilla is thus related to the rapid establishment of a bigger root system that stimulates the general growth of the host plant.

Phenolic compounds are plant secondary metabolites that are biosynthesized through the shikimic acid pathway (Herrmann 1995; Taiz and Zeigler 1998, El-Morsi, *et al*., 2000). Most common classes of plant phenolics having antioxidant properties include those derived from the products of the phenylpropanoid-acetate pathway. Another major property of phenolic compound is antimicrobial activity and it is often assumed that their main role in plants is to act as protective compounds against disease agents such as bacteria, fungi and viruses Candela *et al*., (1995).

3.10 Effect of use three Azotobacter isolates and chemical fertilization on TFs

The concentrations of total flavonoids (TFs) determined in the samples ranged from 56 to 145 µg/100 g fwb in untreated sample and Treatment-4 respectively (Fig. 11). Our results also, showed that the differences between treatment-3, treatment-4 and treatment-6 are not sharp and ranged from 139 to 145 µg/100 g fwb. Flavonoids in tuber plants has many defensive characters, potent antioxidants beside the pharmacologic properties for these reasons, these consider positive indicators in artichoke (Franco *et al*., 2002).

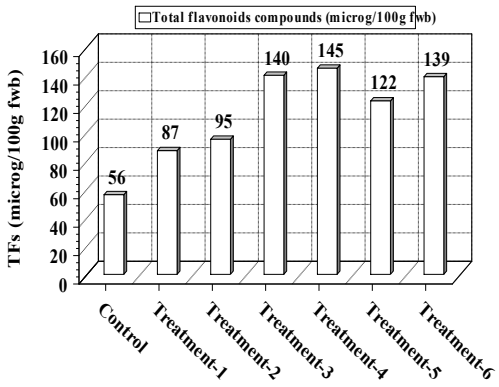

Fig. (10): Effect of biofertilization using three different isolates on total flavonoids compound levels in *Jerusalem artichoke* plants

3-11 Effect of use three Azotobacter isolates and chemical fertilization on TSS

The total soluble sugars (TSS) assayed in the samples ranged from 6.9 to 11.6 % in untreated sample and Treatment-6, respectively (Fig. 11). These results are much lower than those reported by Reust and Dutoit., (1992). Our results also, showed that use of Azotobacter isolates increased the levels of TSS. The crop may be used to produce sugar and fructans, for various usages, such as food, chemical, electronic and pharmaceutical applications (Bosticco *et al*., 1989; ; Marchetti, 1993 ,Baba *et al*., 2006) Jerusalem artichoke accumulates high levels of fructans in their stems and tubers. Fructans and the fructose resulting from fructans hydrolysis can be used in human diet or in medical and industrial applications (Schittenhelm, 1999; Monti *et al*., 2005; Puangbut *et al*., 2012).

It is well documented that Jerusalem artichoke is a viable fructose source. Fructose is more soluble in water than sucrose, so fructose provides a more desirable syrup. In addition, it is 1.5 times sweeter than sucrose and can be consumed safely by diabetics (Mario B , et al ., 2004).

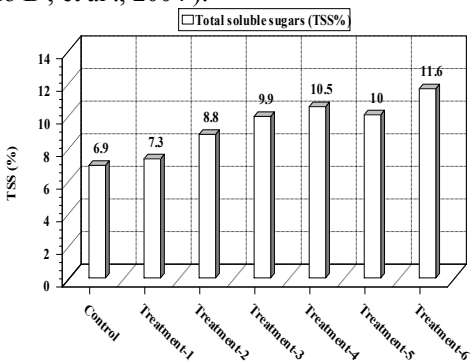

Fig. (11): Effect of biofertilization using three different isolates on total soluble sugars levels in *Jerusalem artichoke* plants.

Reust and Dutoit (1992) found in the *H.tuberosus* trials that a basal fertilizer application of 80 Kg P_2O_5+240 Kg K_2O/ha supplemented by up to 40 Kg N mineral or organic fertilizer at planting, produced tuber yields which ranged from 25.0 to 47.52 t/ha, DM content from 19 to 24%, tuber sugar contents from 11 to 19% and shoot and leaf fresh weight ranged from 5.0 to 48.0 t/ha.

Effect of fertilizer on yield and plant height of *Helianthus tuberosus* Lwas also significant. For example, lack of phosphorus or potassium disturbs tuber morphogenesis, growth and yield, more than aerial growth (Soja *et al*., 1990). The yield response to nitrogen is stronger than to potassium because of the difference in their original content in the soil and because nitrogen determines potential photosynthesis and somewhat increases water use efficiency (Soja and Haunold, 1991). There are some studies on the effect of water and nitrogen on this crop, but these investigations only talk about the effect of water and nitrogen on weight of tubers or aboveground biomass (Long *et al*., 2008) and little talk about the effect of water and nitrogen on the other plant characteristics'.

4. DISCUSSION

In the present work , 8 vegetative characters and 4 chemical constituents of Jerusalem artichoke were studied to through lights on the effect of biofertilization using three different isolates of Azotobacter and two mineral N levels on yield, yield components and some chemical constituents. The reported results indicated that using Azotobacter isolates led to increases in many vegetative plant characteristics and all tested chemical constituents.

In natural conditions plants continually interact with soil microorganisms. This interaction exists primarily at the root level and may be harmful, neutral or beneficial. Regarding the latter, bacteria belonging to the *Azotobacter* and *Azospirillum* genus have been studied as plant growth-promoting rhizobacteria (PGPR; Glick *et al*., 1999). Conceptually, PGPR can affect plant growth and development either directly and indirectly. On the one hand, rhizobacteria may decrease or prevent some effects of phytopathogenic organisms through the production of antibiotics. On the other hand, bacteria either directly provide the plant with different compounds or facilitate their incorporation, like nitrogen fixation or phosphorus solubilization. Production metabolism of phytohormones like auxins, cytokinins and gibberellins (Bottini *et al*., 2004 and Cassan *et al*., 2001) are among the mechanisms used by PGPR to promote plant growth.

The possibility of growing Jerusalem artichoke for energy has attracted the scientific interest in this crop. Portugal imports the main raw materials usually used in other countries to produce bio-ethanol, such as maize and small grains. Portugal also imports sugar derived from cane or sugar beet. Considering that Jerusalem artichoke is a hardy crop that may be cultivated in the new cultivated soil at low cost with simple input techniques and, eventually, without irrigation (Monti *et al*., 2005).

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