

Response of *Senna occidentalis*, Link Plants to Nitrogenous Nutrition its Role in Remediating Some Polluted Soils with Heavy Metals

Shaimaa M.M. Hussein, A. El- Tantawy Abd Allah and H.M. El-Bagoury

Department of Ornamental Horticulture, Faculty of Agriculture, Cairo University, Giza, Egypt

Abstract: This study was carried out in pots at the Experimental Nursery of the Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, during the two successive seasons of 2010 and 2011, with the aim of investigating the response of *Senna occidentalis*, Link plants grown in different soils including soil from Faculty of Agriculture, Cairo University as an un-polluted soil and three different soils polluted with Ni and Zn from Al-Gabal Al-Asfar, Abou Rawash or El-Tebbin to nitrogenous nutrition. Each of ammonium sulphate (20.5 % N) and Cerealin biofertilizer (a commercial product containing *Bacillus polymyxa* and *Azotobacter chroococcum* bacteria), were used as nitrogenous nutrition sources. Nitrogenous treatments included ammonium sulphate at 2 g /pot/month for 2 months and raised to 4 g /pot/month till the end of the experiment (N1) or ammonium sulphate at 4 g /pot/month for 2 months and raised to 8 g /pot/month till the end of the experiment (N2) or biofertilizer only (Bio) or Bio+ N1, Bio+N2 or Bio+0.5 N1, while unfertilized plants were used as a control. In most cases, Al-Gabal Al-Asfar soil was clearly the most effective soil in promoting the growth of plants and increasing their chemical constituents (total chlorophylls, carotenoids contents, total carbohydrates, N, P and K%), followed by the soil of the Faculty of Agriculture. On the other hand, plants grown in El-Tebbin soil gave significantly lower values for vegetative growth characteristics than the plants grown in other soils (in most cases) and the lowest concentrations of the previous chemical constituents, but gave the highest, Ni and Zn concentrations. Nitrogenous fertilization treatments significantly increased vegetative growth characteristics, compared to the control (in most cases). In general, raising the N level resulted in steady significant increases in these parameters and increased the leaf contents of total chlorophylls, carotenoids, total carbohydrates, N, P and K, as well as Ni and Zn uptake, while steadily decreased the extractable Ni and Zn of the used soils. Plants received biofertilizer + N2 treatment gave the highest values followed by plants received N2 treatment and that received biofertilizer + N1 treatment. In many cases, there was no significant difference in vegetative growth of plants received Bio+N2 and that received Bio +N1. Unfertilized control plants gave the lowest values for vegetative growth characteristics and there was no significant difference between unfertilized control plants and that received biofertilizer only in most cases. In general, the plants grown in Al-Gabal Al-Asfar soil and received Bio+N2 gave the highest values for most of the studied vegetative growth characteristics and chemical constituents of the plant, whereas the plants grown in El-Tebbin soil and received no nitrogenous fertilization gave the lowest values. From the above results, it is clear that the best vegetative growth of *Senna occidentalis*, Link plants was obtained when the plants were grown in Al-Gabal Al-Asfar soil and received Bio+N2 treatment. This treatment combination gave the highest values for most of the vegetative growth characteristics and chemical constituents. Generally, Bio+N1 gave vegetative growth insignificantly different than that of the best recorded with plants received Bio+N2. It is also worth mentioning that, in each of the tested soils; applied Bio+N2 gave the best results for most of the studied vegetative growth parameters, as well as for most of the plant chemical components and increased the ability of plants to remediate Ni polluted soils. However, the results showed that *Senna occidentalis*, Link plants cannot remediate Zn polluted soils.

Key words: *Senna occidentalis* • Fertilization • Polluted soils • Heavy metals • Ni • Zn • Hyperaccumulator

INTRODUCTION

Contamination of soils with toxic metals is a frequent global problem. Contaminated sites are to be redeveloped because it is essential to constrain risks to humans, animals and plants. Traditional approaches for estimating such risks are based on the total concentration of each pollutant, but more recently there has been a shift towards the determination of environmentally accepted endpoints, this approach is based on the recognition that the interactions of pollutants with the soil matrix may affect the risk inherent in them [1]. Remediation of metal contaminated soil faces a particular challenge. Unlike organic contaminants, metals cannot be degraded. Commonly, decontamination of metal-contaminated soils requires the removal of toxic metals. The terms tolerant and hyperaccumulator are used when categorizing plants that can grow in the presence of toxic elements. A tolerant species is one that can grow on soil with concentrations of particular elements that are toxic to most other plants; also it is not necessarily an indicator or hyperaccumulator. Hyperaccumulators take up particularly high amounts of a toxic metal. In this respect, many researchers stated that the thresholds for plant hyperaccumulation (shoot dry weight) were set at 1000 mg kg⁻¹ Zn, 100 mg kg⁻¹ Cd, 1000 mg kg⁻¹ Pb. [2, 3].

Recently, phytoextraction, the use of plants to extract toxic metals from contaminated soils, has emerged as an environment-friendly cleanup alternative and as a cost-effective approach because of the high cost cleanup conventional engineering technologies. In addition, because it remediates the soil *in situ*, phytoremediation avoids dramatic landscape disruption and preserves the ecosystem. Phytoremediation is a technology that employs the use of higher plants for the cleanup of contaminated environments. Despite this tremendous potential, phytoremediation is yet to become a commercial technology. Knowledge of basic plant remedial mechanisms in addition to the effect of agronomic practices on these mechanisms are limitations lies within the nature of this novel approach. For example, potential for phytoremediation depends upon the interaction among soil, contaminants, microbes and plants. This complex interaction is affected by a variety of factors, such as climatic conditions and soil properties. Thus, an understanding of the basic plant mechanisms and the effect of agronomic practices on plant/soil/contaminant interaction would allow practitioners to optimize phytoremediation by customizing the process to the site specific conditions [4].

Many researchers stated that some plants belong to genera *Cassia* and *Senna* accumulate heavy metals in their tissues. In this respect, Das *et al.* [5]; Ghosh and Singh [6] and Gupta and Sinha [7] on *Cassia tora*, reported that the plants accumulated high concentrations of Pb, Cu, Ni, Al, Zn, Cd, Fe, Mn and Cr in their leaves and roots. Siringoringo [8] reported that *Cassia multijuga* was found to be highly capable of absorbing and accumulating lead. Dutta and Agrawal [9]; Kumar *et al.* [10] and Raju *et al.* [11] reported that *Cassia siamea* accumulated high concentrations of Ni, Mn, Cr, Pb, Cu, Fe and Zn in leaves and shoots. Also, Hanif *et al.* [12] stated that *Cassia fistula* biomass was very effective for Ni removal from wastewater. Gupta and Sinha [13] reported that *Cassia fistula* played an important role in phytoremediating soils contaminated with K, Fe, Ni, Zn, Mn, Cr, Pb, Cd, Co, Cu and Na. *Cassia siamea* was found to accumulate Fe, Mn, Zn, Cu, Ni, Cr, Pb and Cd at high concentrations and it could be used as a hyperaccumulator plant for bioremediation [14]. Al-Qahtanim [15] indicated that *Cassia italic* is accumulator for Fe, Zn, Cr, Cu, Pb, Ni, Co and Cd.

Senna occidentalis, Link (*Cassia occidentalis*, L.) is a medium-size flowering shrub belonging to family Caesalpiniaceae, which is widespread in warm areas of the world. It reaches heights of about 2 meters and produces yellow flowers in the leaf axils used as a source of colour during the warm months of the year [16]. *Senna occidentalis* has some medicinal uses. It is known as "coffee senna", since its seeds are brewed into a coffee-like beverage for asthma and its flower infusion is used for bronchitis in the Peruvian Amazon. Also, its leaf extracts have exhibited broad-spectrum antibacterial and antifungal activity [17-19], while leaves powders and extracts have proved to be effective in the control of a large variety of insects [20]. Moreover, *Senna occidentalis* plants have also been used to reduce the numbers of mosquitoes indoors [21]. Also, *Senna occidentalis* is used in treatment of liver diseases and its leaf extracts afford significant hepatoprotection [22]. It can therefore be stated that *Senna occidentalis* has an enormous potential use as an ornamental shrub, a medicinal plant and for pest control.

Chemical fertilization improved growth of *Cassia* plants. The nitrogenous nutrition is necessary for the various biochemical processes that occur within the plant and that are essential for plant growth and development [23]. Ramamoorthy and Krishnaveni [24] on *Senna sulfurea*; Hussein [25] and Helmy [26] on *Senna occidentalis*, reported that N fertilization increased plant

height, stem diameter, number of shoots/ plant as well as fresh and dry weights of leaves, stems and roots. They also reported that the total chlorophylls concentrations as well as total carbohydrates, N, P and K in leaves were improved. Kamel and Sakr [27] on *Senna occidentalis*, showed that plant height, stem diameter, number of branches/plant and fresh and dry weights of leaves, stems and roots/plant and dry weight of shoots, total chlorophylls in leaves, total carbohydrates, N, P, K, Cu and Pb in shoots, Cu and Pb uptake as well as extractable Cu and Pb were favourably affected by NPK fertilization. Pratibha [28] pointed out that nitrogen application improved *Cassia angustifolia* yield, Al-Menaie *et al.* [29] revealed that N: P: K at 1g/l gave the highest plant height of *Cassia nodosa* and *Cassia fistula*. In addition, biofertilizers had an enhancing effect on some plant growth characters and this was explained by Subba Rao [30] who attributed their effect to fixing molecular nitrogen, synthesizing and secreting indole acetic acid, cytokinins, gibberellins and cytokinin- or gibberellin-like substances, increasing amino acid content and producing anti-fungal antibiotics which inhibit a variety of soil fungi. Also, Okon [31] mentioned that biofertilizers promote the synthesis of some vitamins, including B12. On the other hand, the beneficial effect of *Bacillus polymyxa* may be attributed to its P-solubilizing action [30]. A different explanation to the mode of action of *B. polymyxa* was proposed by Gouzou *et al.* [32] who reported that inoculation of the soil with *B. polymyxa* caused an increase in aggregated soil particles by 57% which led to a more porous structure within the rhizosphere soil and, consequently, enhanced water retention and nutrient transfer in the rhizosphere of the plant.

This study was conducted with the aim of investigating the response of *Senna occidentalis*, Link plants to nitrogenous nutrition and their role in remediating different polluted soils with heavy metals.

MATERIALS AND METHODS

This study was carried out at the Experimental Nursery of the Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza, during the two successive seasons of 2010 and 2011, with the aim of investigating the response of *Senna occidentalis*, Link, L. plants to chemical and biofertilizer nitrogenous nutrition and their role in remediating different polluted soils with heavy metals.

Seeds of *Senna occidentalis* were sown on 1st March 2010 and 2011 (in the first and second seasons, respectively), in a glasshouse in 8-cm plastic pots filled

with a 1:1 (v/v) mixture of sand and clay. On 1st May 2010 and 2011 (in the two seasons, respectively), the seedlings (12 cm) were transplanted individually into plastic pots (30-cm diameter) filled with 5 kg of: (1) clay loam soil from the nursery of the Ornamental Horticulture Department, Faculty of Agriculture, Cairo University, Giza (as an unpolluted soil), or one of three different polluted soils including: (2) sandy loam soil from Al-Gabal Al-Asfar, Cairo, (3) sandy loam soil from Abou Rawash, Giza, or (4) clay soil from El-Tebbin, Cairo. The source of pollutants, general characteristics, available macronutrients and DTPA extractable nickel and zinc were determined in surface samples (0-30 cm) of the four studied soils and are shown in Table 1. The pots were kept outdoors throughout the experiment.

After 15 days from transplanting, chemical nitrogenous fertilizer was applied to the plants at the rates of 2 g ammonium sulphate (20.5 % N) /pot/month for 2 months and raised to 4 g /pot/month till the end of the experiment (N1) or 4 g ammonium sulphate (20.5 % N) /pot/month for 2 months and raised to 8 g /pot/month till the end of the experiment (N2). The commercial biofertilizer "Cerealin" [produced by Egyptian Ministry of Agriculture and containing *Bacillus polymyxa* and *Azotobacter chroococcum* bacteria, each at 10⁷ colony forming units (CFU)/ml] were used singly as a biofertilizer (Bio) or in different combinations with chemical N fertilizer (Bio+N1, Bio+N2 and Bio+0.5 N1). With biofertilization treatments, 20 ml of the biofertilizer was injected in a hole adjacent to the plant roots, also additional boost (ten ml of the inoculum) was injected in a hole adjacent to the plants monthly. An unfertilized control was also included in the experiment.

All pots received phosphorus fertilizer in the form of calcium superphosphate (15.5% P₂O₅), which was mixed into the soil two weeks before transplanting the seedlings, at the rate of 10 g/pot. Potassium fertilizer was added in the form of potassium sulphate (48% K₂O) at the rate of 9 g/pot, divided into three equal doses, which were applied after 2, 10 and 18 weeks from transplanting.

The plants were irrigated every 3 days using a tap water (with a total salts concentration of 270 ppm). At each irrigation, the plants were watered till 90% of soil field capacity (F.C.). The soil moisture tension was measured before each irrigation using digital microtensiometers and the quantity of water needed to reach 90% F.C. was calculated, as described by Richards [33].

The layout of the experiment was a split-plot design, with the main plots arranged in a randomized complete blocks design, with three blocks (replicates). The main

Table 1: Location, source of pollutants, general characteristics, available macronutrients and DTPA extractable nickel and zinc of the studied soils

No.	Location	Source of pollutant*	PH (1: 2.5) (dS/m)	E.C. (1: 2.5)	Organic matter (%)	CEC (meq/100g)	CaCO ₃ %	Particle size distribution					Field capacity (%)	Available macronutrients (ppm)			DTPA extractable (µg/ g)	
								Coarse sand (%)	Fine sand (%)	Silt (%)	Clay (%)	Texture class		N	P	K	Ni	Zn
1	Fac. Agric. Giza	Nil	7.9	0.47	1.4	31	0.9	6.96	18.45	40.37	34.22	Clay loam	33	18	17	239	0.2	7.0
2	Al-Gabal Al-Asfar	D and I	6.8	1.07	3.8	43	1.0	32.44	37.89	11.74	17.93	Sandy loam	25	80	31	442	12.1	91.0
3	Abou Rawash	D and I	7.1	0.91	2.5	12	2.8	32.02	39.45	12.69	15.84	Sandy loam	23	57	25	330	12.3	95.0
4	El-Tebbin	I	7.8	0.62	0.7	42	4.9	10.23	19.88	25.75	44.14	Clay	38	50	16	323	13.2	98.0

*D: Domestic wastes. I: Industrial wastes

plots were assigned to the different soils, while the sub-plots were assigned to the fertilization treatments. The study included 28 treatments [4 different soils × 7 fertilization treatments (including the control)], with each block consisting of 84 plants (3 plants/ treatment).

At the termination of each season (on 30th December 2010 and 2011 in the first and second seasons, respectively), plants were cut 5 cm above the soil surface, rinsed once with diluted HCl and twice with H₂O. Data were recorded on some plant vegetative growth characteristics, including plant height (cm), stem diameter (mm) at a height of 5 cm from the soil surface, number of branches/plant, as well as the fresh and dry weights of shoots, leaves and roots/plant. The data on the vegetative growth characteristics were subjected to statistical analysis of variance and the means were compared using the "Least Significant Difference (L.S.D.)" test at the 5% level, as described by Little and Hills [34].

Chemical analysis of fresh leaf samples was also conducted to determine their concentrations of chlorophyll a, b and carotenoids (using the method described by Moran [35]). After that total chlorophylls (a+b) was calculated. In addition, samples of leaves were oven-dried at a temperature of 70°C for 24 hours and their contents of nutrients were extracted using the method described by Piper [36]. The nutrients extract was chemically analyzed to determine the percentages of nitrogen [37], phosphorus [38] potassium (estimated photometrically using a Jenway flamephotometer), nickel and zinc using an atomic absorption spectrophotometer, model GBC, 932AA). Also the concentration of total carbohydrates in dried leaf samples was determined using the method described by Dubois *et al.* [39]. The uptake of Ni and Zn was calculated by multiplying the dry weight of above-ground parts of the plant by the concentration of the two minerals in the plant tissues.

At the end of the experiment, the soils were analyzed for DTPA- extractable Ni and Zn, as recommended by Lindsay and Norvell [40].

RESULTS AND DISCUSSION

Vegetative Growth

Effect of Different Soils: Regarding the effect of different soils on vegetative growth characteristics (plant height, stem diameter, number of shoots/ plant, root length as well as fresh and dry weights of leaves, shoots and roots) of *Senna occidentalis*, Link plants, regardless of the effect of fertilization treatments, the results recorded in the two seasons (Tables 2-4) showed that among the different soils used, the sandy loam soil obtained from Al-Gabal Al-Asfar was clearly the most effective one for promoting the vegetative growth characteristics of *Senna occidentalis*, Link plants, giving the significantly higher values, followed by plants grown in Faculty of Agriculture (clay loam) soil, loam soil from Abou Rawash and clay soil from El-Tebbin. Two exceptions to this general trend were recorded since plants grown in Faculty of Agriculture soil gave insignificantly higher root length in the second season and insignificantly fresh weight of shoots in the first season than that recorded with plants grown in Al-Gabal Al-Asfar soil. In both seasons, there were no significant difference in the plant height, fresh weights of leaves, shoots and roots of plants grown in soil obtained from Al-Gabal Al-Asfar and that grown in soil obtained from Faculty of Agriculture. The plants grown in El-Tebbin soil gave the significantly lower values for vegetative growth characteristics, as compared to plants grown in most of other soils.

The increase in vegetative growth of plants grown in Al-Gabal Al-Asfar soil may be attributed to its high content of organic matter (compared to other soils used) which has favorable effects on the soil, such as improving some of its physiochemical properties, preventing salt injury to plants that sometimes results from concentration of chemical fertilizers through the buffering properties of organic matter and providing soil with the essential macro and micronutrients [41, 42].

Table 2: Effect of different soils as well as bio- and chemical N fertilizers on plant height (cm), stem diameter (mm), number of shoots/plant and root length (cm) of *Senna occidentalis*, Link. plants during 2010 and 2011 seasons

Fertilization treatments (F)	First season (2010)					Second season (2011)				
	Soils (S)					Soils (S)				
	S1	S2	S3	S4	Mean	S1	S2	S3	S4	Mean
Plant height (cm)										
Control	84.5	95.7	78.5	70.2	82.2	115.2	117.5	103.5	101.5	109.4
N1	99.7	106.7	93.4	82.5	95.6	126.9	134.5	118.2	112.4	123.0
N2	109.2	103.5	99.7	89.4	100.5	140.8	143.6	130.7	116.4	132.9
Bio	94.7	97.9	86.4	72.1	87.8	121.9	125.6	110.1	105.8	115.9
Bio + N1	102.8	110.3	95.9	85.7	98.7	130.5	140.5	126.6	115.9	128.4
Bio + N2	111.3	119.4	101.2	94.5	106.6	144.5	150.7	134.1	123.7	138.3
Bio + 0.5 N1	99.2	103.5	89.5	80.7	93.2	125.7	128.1	115.2	107.2	119.1
Mean	100.2	105.3	92.1	82.2	----	129.4	134.4	119.8	111.8	----
LSD 0.05	S		6.9			S		8.5		
	F		8.3			F		12.4		
	Sx F		10.2			Sx F		15.6		
Stem diameter (mm)										
Control	10.1	10.1	7.6	7.1	8.7	10.9	13.1	10.1	8.3	10.6
N1	13.7	13.9	10.4	9.2	11.8	18.1	19.9	15.2	13.1	16.6
N2	18.1	20.1	14.2	13.1	16.4	20.1	20.8	15.5	14.7	17.8
Bio	10.8	9.8	8.5	7.4	9.1	12.8	15.6	10.9	8.9	12.1
Bio + N1	17.9	19.5	13.5	10.1	15.3	19.3	20.4	15.8	13.6	17.3
Bio + N2	20.4	20.3	15.6	13.8	17.5	20.1	21.5	14.6	15.1	17.8
Bio + 0.5 N1	12.3	13.1	9.8	8.3	10.9	13.7	19.8	11.3	10.2	13.8
Mean	14.8	15.3	11.4	9.9	----	16.4	18.7	13.3	12.0	----
LSD 0.05	S		1.1			S		1.4		
	F		1.8			F		1.9		
	Sx F		2.3			Sx F		2.8		
Number of shoots/plant										
Control	8.2	8.8	5.8	5.2	7.0	8.8	9.8	7.7	6.3	8.2
N1	10.7	12.7	10.7	7.3	10.4	10.7	10.8	9.8	9.2	10.1
N2	12.2	14.8	11.8	9.8	12.2	12.3	12.8	11.7	11.3	12.0
Bio	9.7	10.3	6.2	5.7	8.0	9.2	10.3	8.8	7.8	9.0
Bio + N1	11.8	14.2	11.3	9.3	11.7	11.2	11.3	10.2	9.8	10.6
Bio + N2	12.8	15.7	12.2	10.3	12.8	12.8	13.3	12.8	12.2	12.8
Bio + 0.5 N1	10.2	11.8	8.8	6.8	9.4	9.7	10.7	9.2	8.7	9.6
Mean	10.8	12.6	9.5	7.8	----	10.7	11.3	10.0	9.3	----
LSD 0.05	S		1.4			S		1.6		
	F		1.8			F		2.3		
	Sx F		2.3			Sx F		2.5		
Root length (cm)										
Control	30.9	32.3	24.1	20.5	27.0	30.8	28.6	24.8	23.1	26.8
N1	40.6	46.5	30.5	28.6	36.6	40.5	35.8	34.9	33.6	36.2
N2	45.5	56.5	41.5	34.5	44.5	50.8	50.9	40.5	38.6	45.2
Bio	34.5	38.9	25.9	22.8	30.5	33.9	36.9	29.6	24.6	31.3
Bio + N1	42.5	49.3	36.1	30.9	39.7	46.8	48.5	39.5	34.9	42.4
Bio + N2	48.9	57.1	43.1	36.1	46.3	51.2	54.3	43.6	39.9	47.3
Bio + 0.5 N1	35.9	36.5	28.9	23.9	31.3	38.4	32.5	30.8	28.8	32.6
Mean	39.8	45.3	32.9	28.2	----	41.8	41.1	34.8	31.9	----
LSD 0.05	S		2.8			S		2.5		
	F		3.4			F		4.5		
	Sx F		5.1			Sx F		5.9		

S1: Fac. Agric., Giza. S2: Al – Gabal Al- Asfar. S3: Abou -Rawash. S4: El-Tebbin

N1 = 2 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 4 g /pot till the end of the experiment

N2 = 4 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 8 g /pot till the end of the experiment

Bio= "Cerealin" containing *Bacillus polymyxa* and *Azotobacter chroococcum* bacteria

Table 3: Effect of different soils as well as bio- and chemical N fertilizers on fresh weights of leaves, shoots and roots as well as dry weights of leaves/ plant (g) of *Senna occidentalis*, Link. plants during 2010 and 2011 seasons

Fertilization treatments (F)	First season (2010)					Second season (2011)				
	Soils (S)					Soils (S)				
	S1	S2	S3	S4	Mean	S1	S2	S3	S4	Mean
Fresh weight of leaves/plant (g)										
Control	80.6	86.1	70.8	62.3	75.0	92.9	101.6	79.5	64.5	84.6
N1	125.6	115.9	111.4	79.5	108.1	131.4	135.4	115.6	83.6	116.5
N2	134.1	132.5	122.8	98.1	121.9	166.4	176.8	132.1	111.5	146.7
Bio	85.2	90.5	78.5	68.5	80.7	100.4	120.8	81.9	71.8	93.7
Bio + N1	130.8	120.4	115.6	94.4	115.3	150.5	156.9	131.5	99.7	134.7
Bio + N2	135.8	135.8	131.5	105.1	127.1	170.8	178.5	136.7	115.8	150.5
Bio + 0.5 N1	94.6	110.6	89.1	70.1	91.1	118.6	131.6	83.7	75.9	102.5
Mean	112.4	113.1	102.8	82.6	----	133.0	143.1	108.7	89.0	----
LSD 0.05	S		9.4					13.2		
	F		12.1					19.5		
	Sx F		16.8					28.5		
Fresh weight of shoots/plant (g)										
Control	140.6	152.8	100.5	92.5	121.6	138.5	150.4	112.4	98.1	124.9
N1	191.7	180.4	149.5	122.8	161.1	176.4	184.3	141.5	134.0	159.1
N2	229.5	220.5	175.4	151.5	194.2	199.5	211.4	160.8	144.8	179.1
Bio	169.5	165.4	121.5	101.5	139.5	146.1	169.1	116.9	115.4	136.9
Bio + N1	220.5	210.6	170.5	148.5	187.5	184.5	196.5	154.5	142.5	169.5
Bio + N2	236.8	230.1	196.1	170.1	208.3	207.3	213.5	169.5	150.2	185.1
Bio + 0.5 N1	182.4	170.6	128.4	116.8	149.6	165.1	168.5	126.8	120.7	145.3
Mean	195.9	190.1	148.8	129.1	----	173.9	184.8	140.3	129.4	----
LSD 0.05	S		14.9					16.8		
	F		21.6					23.8		
	Sx F		26.7					29.6		
Fresh weight of roots/plant (g)										
Control	90.5	84.5	76.9	64.1	79.0	115.7	119.5	70.6	60.1	91.5
N1	135.1	139.8	111.1	94.5	120.1	134.5	140.5	119.5	101.4	124.0
N2	146.8	159.5	136.5	115.8	139.7	150.6	161.4	135.4	111.8	139.8
Bio	108.5	105.1	80.5	72.2	91.6	119.8	125.6	80.1	75.9	100.4
Bio + N1	140.6	145.1	118.9	99.7	126.1	148.1	156.9	122.8	105.8	133.4
Bio + N2	154.1	162.1	140.5	131.5	147.1	159.4	163.8	140.7	122.9	146.7
Bio + 0.5 N1	115.4	126.1	95.8	86.1	105.9	129.6	131.7	106.5	93.8	115.4
Mean	127.3	131.7	108.6	94.8	----	136.8	142.8	110.8	96.0	----
LSD 0.05	S		11.6					10.5		
	F		18.9					16.4		
	Sx F		24.6					25.9		
Dry weight of leaves/plant (g)										
Control	12.6	14.1	11.3	9.8	12.0	15.1	17.7	12.0	9.9	13.7
N1	23.2	22.3	18.3	13.2	19.3	22.5	26.3	19.1	13.5	20.4
N2	26.3	28.4	21.4	16.9	23.3	30.1	34.9	24.2	18.6	27.0
Bio	14.3	15.2	12.8	11.0	13.3	17.0	21.1	13.4	11.1	15.7
Bio + N1	24.9	24.2	19.5	15.5	21.0	25.8	31.0	22.1	16.1	23.8
Bio + N2	27.1	28.2	23.6	18.6	24.4	32.4	36.1	25.1	19.7	28.3
Bio + 0.5 N1	16.8	20.6	14.7	11.5	15.9	19.2	23.9	13.8	11.7	17.2
Mean	20.7	21.9	17.4	13.8	----	23.2	27.3	18.5	14.4	----
LSD 0.05	S		1.9					2.5		
	F		3.5					3.5		
	Sx F		4.6					5.3		

S1: Fac. Agric., Giza.

S2: Al – Gabal Al- Asfar.

S3: Abou -Rawash.

S4: El-Tebbin

N1 = 2 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 4 g /pot till the end of the experiment

N2 = 4 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 8 g /pot till the end of the experiment

Bio= "Cerealin" containing *Bacillus polymyxa* and *Azotobacter chroococcum* bacteria

Table 4: Effect of different soils as well as bio- and chemical N fertilizers on dry weights of shoots and roots/plant (g) as well as total chlorophylls and carotenoids concentrations (mg/g fresh matter) of *Senna occidentalis*, Link. plants during 2010 and 2011 seasons

Fertilization treatments (F)	First season (2010)					Second season (2011)				
	Soils (S)					Soils (S)				
	S1	S2	S3	S4	Mean	S1	S2	S3	S4	Mean
Dry weight of shoots/plant (g)										
Control	26.0	30.1	17.7	16.3	22.5	24.6	30.3	20.1	16.1	22.8
N1	37.7	39.7	28.6	23.0	32.3	37.1	41.6	26.9	23.4	32.3
N2	47.7	51.8	36.7	29.6	41.5	44.1	50.4	32.2	26.8	38.4
Bio	32.3	33.1	22.5	18.4	26.6	27.4	34.0	21.6	19.3	25.6
Bio + N1	43.5	49.3	33.2	27.7	38.4	39.8	46.4	30.0	25.1	35.3
Bio + N2	53.6	55.2	41.0	34.0	46.0	46.6	52.9	34.9	28.6	40.8
Bio + 0.5 N1	33.9	35.4	24.0	21.3	28.7	32.7	37.1	23.8	20.6	28.6
Mean	39.2	42.1	29.1	24.3	---	36.0	41.8	27.1	22.8	---
LSD 0.05	S		3.2					3.4		
	F		3.8					5.4		
	Sx F		4.3					6.9		
Dry weight of roots/plant (g)										
Control	18.8	18.6	15.1	12.1	16.2	25.1	28.2	13.7	11.2	19.6
N1	35.7	41.0	25.4	21.0	30.8	32.8	39.2	27.7	21.9	30.4
N2	42.9	50.9	35.1	28.7	39.4	39.8	46.7	35.5	26.2	37.1
Bio	24.5	24.1	17.5	15.0	20.3	27.8	30.8	17.4	15.0	22.8
Bio + N1	41.2	41.5	28.2	22.4	33.3	36.9	45.5	29.7	23.7	34.0
Bio + N2	47.5	51.7	36.0	32.5	41.9	44.3	48.8	37.3	29.6	40.0
Bio + 0.5 N1	28.2	33.1	20.4	17.5	24.8	29.5	34.1	23.6	18.9	26.5
Mean	34.1	37.3	25.4	21.3	---	33.7	39.0	26.4	20.9	---
LSD 0.05	S		3.5					3.2		
	F		4.9					4.6		
	Sx F		6.8					6.2		
Total chlorophyll (a+b) concentration (mg/g fresh matter)										
Control	0.98	1.07	0.91	0.84	0.95	1.19	1.26	0.96	0.85	1.07
N1	1.20	1.31	1.18	0.97	1.17	1.4	1.53	1.15	1.05	1.28
N2	1.35	1.47	1.33	1.07	1.31	1.54	1.73	1.36	1.14	1.44
Bio	1.05	1.16	0.98	0.87	1.02	1.25	1.32	1.01	0.9	1.12
Bio + N1	1.26	1.36	1.24	1.03	1.22	1.5	1.56	1.24	1.14	1.36
Bio + N2	1.48	1.52	1.36	1.17	1.38	1.6	1.76	1.35	1.22	1.48
Bio + 0.5 N1	1.13	1.19	1.10	0.93	1.09	1.3	1.5	1.09	0.99	1.22
Mean	1.21	1.30	1.16	0.98	---	1.40	1.52	1.17	1.04	---
Carotenoids concentration (mg/g fresh matter)										
Control	0.30	0.35	0.30	0.23	0.30	0.22	0.241	0.22	0.17	0.21
N1	0.45	0.48	0.40	0.32	0.41	0.27	0.28	0.23	0.22	0.25
N2	0.52	0.56	0.52	0.42	0.51	0.29	0.31	0.28	0.26	0.29
Bio	0.32	0.40	0.32	0.28	0.33	0.23	0.26	0.22	0.21	0.23
Bio + N1	0.47	0.50	0.49	0.38	0.46	0.29	0.29	0.27	0.25	0.28
Bio + N2	0.63	0.59	0.50	0.49	0.55	0.32	0.32	0.26	0.27	0.29
Bio + 0.5 N1	0.43	0.42	0.38	0.28	0.38	0.28	0.27	0.23	0.20	0.25
Mean	0.45	0.47	0.42	0.34	---	0.27	0.28	0.24	0.23	---
Total carbohydrates (% D.W. of leaves)										
Control	27.5	28.0	24.1	22.5	25.5	25.7	28.5	22.8	21.3	24.6
N1	29.8	30.5	26.8	24.8	28.0	27.8	30.8	25.7	23.4	26.9
N2	32.4	32.8	28.2	25.7	29.8	28.9	33.7	26.7	24.9	28.6
Bio	27.8	28.4	24.8	22.9	26.0	25.9	28.9	23.4	21.6	25.0
Bio + N1	30.1	31.9	26.9	24.9	28.5	28.5	31.5	25.9	23.9	27.5
Bio + N2	32.8	34.8	28.9	25.8	30.6	30.2	34.2	27.9	26.7	29.8
Bio + 0.5 N1	28.5	28.9	25.1	23.7	26.6	27.1	30.1	24.5	22.9	26.2
Mean	29.8	30.8	26.4	24.3	---	27.7	31.1	25.3	23.5	---

S1: Fac. Agric., Giza.

S2: Al – Gabal Al- Asfar.

S3: Abou -Rawash.

S4: El-Tebbin

N1 = 2 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 4 g /pot till the end of the experiment

N2 = 4 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 8 g /pot till the end of the experiment

Bio= "Cerealin" containing *Bacillus polymyxa* and *Azotobacter chroococcum* bacteria

Effect of Fertilization Treatments: Concerning the effect of fertilization treatments on vegetative growth characteristics of *Senna occidentalis*, Link plants, regardless of the effect of different soils, the results recorded in the two seasons (Tables 2-4) showed that the vegetative growth characteristics of *Senna occidentalis*, Link plants was favorably affected by the different fertilization treatments. In most cases, plants receiving any of the tested fertilization treatments gave significantly higher values, compared to unfertilized (control) plants. In both seasons, plants received biofertilizer + N2 treatment gave the highest values followed by plants received N2 treatment and that received biofertilizer + N1 treatment. In both seasons, there was no significant difference in the plant height, number of shoots/plant as well as fresh weights of leaves and shoots/plant recorded with plants received Bio+N2, N2 and Bio+N1 treatments. The treatments of Bio+N1 and Bio+N2 insignificantly improved vegetative growth characteristics as compared to the same treatments without biofertilization, in most cases. In both seasons, unfertilized control plants gave the lowest values for vegetative growth characteristics and there was no significant difference between unfertilized control plants and that received biofertilizer only in most cases. Results are in agreement with perior studies [25-27, 43, 44].

The favorable effect of the different N fertilization treatments on the vegetative growth characteristics (compared to the control) can be attributed to the important role played by N in the physiological processes within the plant, which in turn affect the growth of the vegetative and root systems. Also, nitrogen is present in the structure of protein molecules. Besides, N is present in coenzymes which are essential to the function of many enzymes that play roles in the synthesis of all metabolic intermediates, cellular structure components and storage components which constitute the plant body and are required for the meristematic activity and growth of cells and organs. N is found in cytochromes which are a major part of the chlorophyll molecule and is therefore necessary for photosynthesis and respiration [24, 45]. The positive effect of bio-fertilization on vegetative growth can be explained, at least in part, by that azotobacter species are plant growth-promotive bacteria whose beneficial effects have been postulated to influence the hormonal balance of the plant and to secrete phytohormones [e.g. indole-3-acetic acid (IAA), cytokinins and gibberellins] which could stimulate plant growth, absorption of nutrients and photosynthesis process. Also, phosphobacteria improve plant growth due

to biosynthesis of plant growth substances rather than their action in solubilizing inorganic phosphate by secreting phosphatase enzyme and liberating phosphorous from organic compounds which make phosphorus available to the plants [46-52].

Interaction Between the Effects of Different Soils and Nitrogenous Nutrition: Concerning the interaction between the effects of different soils and nitrogenous nutrition, data in Tables (2-4) showed that in both seasons, the *Senna occidentalis*, Link plants grown in Al-Gabal Al-Asfar soil and fertilized with Bio +N2 treatment gave the significantly higher vegetative growth characteristics values as compared to most of the other treatments, in most cases. In both seasons, in most cases, there was no significant difference between the values of different vegetative growth characteristics of plants grown in Al-Gabal Al-Asfar soil and received Bio+N2 treatment or that grown in the same soil and received Bio+N1 treatment.

Effect on the Chemical Composition of the Plant Total Chlorophylls (a+b) and Carotenoids Concentrations: Data presented in Table 4 revealed that, among the different soils used, Al-Gabal Al-Asfar soil was the most effective soil for increasing total chlorophylls (a+b) (1.30 and 1.52 mg/g fresh matter in both seasons, respectively) and carotenoids (0.47 and 0.28 mg/g fresh matter in the first and second seasons, respectively) in the leaves, followed by Faculty of Agriculture, Abou Rawash soils. On the other hand, the lowest total chlorophylls (a+b) (0.98 and 1.04 mg/g fresh matter in both seasons, respectively) and carotenoids (0.34 and 0.23 mg/g fresh matter in both seasons, respectively) in the leaves were found in the plants grown in El-Tebbin soil, which contained the highest Ni and Zn concentrations. Such results are in agreement with the findings on *Tagetes minuta*, *T. patula* and *T. erecta* [53], *Gazania splendens* and *Vinca rosea* [54] and *Brassaia arboricola* var. *variegata* and *Ficus microcarpa* var. *Hawaii* [55]. Also, Hussein and Kamel [56] on *Amaranthus*, found that Al-Gabal Al-Asfar soil was the most effective soil in increasing total chlorophylls and carotenoids followed by the soil of the Faculty of Agriculture.

The data presented in Table 4 showed that N fertilization had a favourable effect on total chlorophylls (a+b) and carotenoids synthesis and accumulation in the leaves of *Senna occidentalis*, Link plants. In both seasons, plants receiving the different N fertilization treatments had higher concentration of total chlorophylls

(a+b) and carotenoids than unfertilized (control) plants. Moreover, the total chlorophylls (a+b) and carotenoids concentrations were steadily increased with raising the nitrogen level. Generally, plants received Bio+N2 gave the highest total chlorophylls (a+b) (1.38 and 1.48 mg/g fresh matter in both seasons, respectively) and carotenoids (0.55 and 0.29 mg/g fresh matter in both seasons, respectively) followed by N2, Bio+N1, N1, Bio+0.5N1 and Bio treatments. The increase in total chlorophylls (a+b) concentrations can be attributed to the role played by nitrogen as an essential component in the structure of porphyrines, which are found in many metabolically active compounds, including chlorophylls. Chlorophylls are bound to and perhaps even embedded within protein molecules [45]. These results are in agreement with that findings on *Senna occidentalis* [27], *Tagetes patula* [57], *Tagetes erecta* and *Amaranthus tricolor* [58], *Senna sulfurea* [59], *Hibiscus sabdariffa* [60], *Capsicum annum* [61] and *Anethum graveolens* [62]. Also, in most cases, the treatments of Bio+N1 and Bio+N2 improved total chlorophylls (a+b) and carotenoids concentrations as compared to the same treatments without biofertilization. Such results are in agreement with Mazrou [61] on *Cymbopogon citratus* in which *Azotobacter chroococcum*, *Bacillus megatherium* or *Trichoderma reesei* individually or in combinations increased chlorophylls and carotenoids contents in the leaves in comparison with the un-inoculated plants. Unfertilized control plants gave the lowest total chlorophylls (a+b) (0.95 and 1.07 mg/g fresh matter in both seasons, respectively) and carotenoids concentrations (0.30 and 0.21 mg/g fresh matter in both seasons, respectively).

Concerning the interaction between the effects of nitrogenous nutrition and different soils, data in Table 4 showed that, in both seasons, leaves of *Senna occidentalis*, Link plants grown in Al-Gabal Al-Asfar soil and received Bio+N2 gave the highest values of total chlorophylls (a+b) and carotenoids concentrations in most cases. In both seasons, the leaves of plants grown in El-Tebbin soil without fertilization gave the lowest total chlorophylls (a+b) and carotenoids concentrations.

Total Carbohydrates Content: Data presented in Table 4 revealed that in both seasons, among the different soils used, Al-Gabal Al-Asfar soil was the most effective soil for increasing total carbohydrates concentration (30.8 and 31.1%, in the first and second season, respectively) in the leaves, followed by Faculty of Agriculture (29.8 and 27.7 %, in both seasons, respectively), Abou Rawash soils (26.4 and 25.3%, in both

seasons, respectively). On the other hand, the lowest total carbohydrates concentration (24.3 and 23.5 %, in both seasons, respectively) in the leaves was found in the plants grown in El-Tebbin soil. The favorable effect of Al-Gabal Al-Asfar soil on carbohydrate synthesis and accumulation may be attributed to its high cation exchange capacity which allows the plant roots to take up the macro and micronutrients including the potassium needed for activation of the enzymes necessary for photosynthesis and, consequently, the synthesis of carbohydrates. On the other hand, the unfavorable effect of the El-Tebbin soil on the total carbohydrates content in the leaves (in both seasons) can be easily explained since, as previously mentioned, the El-Tebbin soil has high concentrations of Ni and Zn, which lead to a reduction in the content of chlorophylls. This reduction in the contents of chlorophylls results in a reduction in the rate of photosynthesis and a reduction in carbohydrate synthesis and accumulation [45]. In this respect Hussein and Kamel [56] on *Amaranthus*, reported that Al-Gabal Al-Asfar soil was the most effective soil in increasing total carbohydrates followed by the soil of the Faculty of Agriculture.

Data presented in Table 4 showed that N fertilization had a favorable effect on total carbohydrates synthesis and accumulation in the leaves of *Senna occidentalis*, Link plants. In both seasons, plants receiving the different N fertilization treatments had higher concentrations of total carbohydrates than unfertilized (control) plants. Moreover, the total carbohydrates concentrations were steadily increased with raising the nitrogen level. Also, the treatments of Bio+N1 and Bio+N2 improved total carbohydrates concentrations as compared to the same treatments without biofertilization. Plants received Bio+N2 gave the highest total carbohydrates concentrations in leaves (30.6 and 29.8 %, in both seasons, respectively) followed by N2, Bio+N1, N1, Bio+0.5N1 and Bio treatments. In the first season leaves of plants received Bio+0.5N1 gave the same concentration of total carbohydrates recorded in plants received biofertilization only. Unfertilized control plants gave the lowest total carbohydrates concentration in leaves (25.5 and 24.6 %, in both seasons, respectively). These results can be easily explained by the indirect effect of nitrogenous fertilization on carbohydrate synthesis. As previously mentioned the nitrogen supplied by fertilization is essential in the structure of porphyrines and, consequently, leads to an increase in the content of chlorophylls. Also, the porphyrine molecules are found in the cytochrome enzymes essential in photosynthesis. This increase in the

contents of chlorophylls and cytochrome enzymes results in an increase in the rate of photosynthesis and a promotion in carbohydrate synthesis and accumulation [45]. Such results are in agreement with that reported on *Senna occidentalis* [27], *Tagetes erecta* and *Amaranthus tricolor* [58], *Senna sulfurea* [59], *Hibiscus sabdariffa* [60] and *Catharanthus roseus* [64].

Concerning the interaction between the effects of nitrogenous nutrition and different soils, data in Table 4 showed that, in both seasons, leaves of *Senna occidentalis*, Link plants grown in Al-Gabal Al-Asfar soils and received Bio+N2 gave the highest values of total carbohydrates concentration (34.8 and 34.2 %, in both seasons, respectively). In both seasons, the leaves of plants grown in El-Tebbin soil without fertilization gave the lowest total carbohydrates concentration (22.5 and 21.3% in both seasons, respectively).

N, P and K % in Leaves: Data presented in Table 5 showed that, in the first season, *Senna occidentalis* plants grown in Al-Gabal Al-Asfar soil had the highest N, P and K% (1.62, 0.14 and 1.70 % for N, P and K, respectively), followed by those grown in the Faculty of Agriculture soil (1.55, 0.13 and 1.62 %, respectively) and Abou Rawash soil (1.36, 0.10 and 1.36 %, respectively), in this order, whereas in the second season plants grown in Al-Gabal Al-Asfar soil had the highest N, P and K % (1.45, 0.11 and 1.48 %, respectively), followed by those grown in Abou Rawash soil (1.38, 0.10 and 1.41 %, respectively) and Faculty of Agriculture soil (1.35, 0.10 and 1.36 %, respectively). Plants grown in Faculty of Agriculture soil gave the same P% recorded in the leaves of plants grown in Abou Rawash soil (0.10%). In both seasons, plants grown in El-Tebbin soil gave the lowest N, P and K % (1.24, 0.08 and 1.21% for N, P and K in the first season and 1.32, 0.09 and 1.31%, respectively in the second one). These lowest N, P and K % that were recorded in plants grown in El-Tebbin soil are associated with the highest content of heavy metals (Ni and Zn) in this soil (Table 1). Such results are in agreement with that recorded on *Casuarina glauca*, *Taxodium distichum* and *Populus nigra* [65].

Data presented in Table 5 showed that N fertilization had a favorable effect on N, P and K % in the leaves of *Senna occidentalis*, Link plants. In both seasons, in most cases, plants receiving the different N fertilization treatments had higher N, P and K % than unfertilized (control) plants. The only exception to this trend was recorded in the second season with plants received

biofertilization only which gave P% such that recorded with unfertilized control plants. Moreover, N, P and K % were steadily increased with raising the nitrogen level, in most cases. The exception to this trend was recorded in the second season with plants received N2, Bio+N1 or Bio+N2 which gave the same P%. Also, the treatments of Bio+N1 and Bio+N2 improved N, P and K % as compared to the same treatments without biofertilization, in most cases. In the first season, plants received Bio+N2 gave the highest N, P and K % in leaves (1.66, 0.15 and 1.75%, respectively). In the second season, plants received N2 gave the highest N% (1.52%) and K% (1.58%), whereas the highest P% (0.12%) was recorded with plants received N2, Bio+N1 and N2. Unfertilized control plants gave the lowest N (1.19 and 1.21% in the first and second seasons, respectively), P (0.07 and 0.08%, respectively) and K % (1.14 and 1.18%, respectively) in leaves. The increase in the percentage of N, P and K% was explained by Jain [66] who stated that raising the level of N in the root medium leads to an increase in vegetative growth and this may be accompanied by an increase in the absorption of this essential element. These results are in agreement with that reported on *Senna occidentalis* [27], *Tagetes erecta* and *Amaranthus tricolor* [58], *Senna sulfurea* [59], *Hibiscus sabdariffa* [60], *Anethum graveolens* [62], *Catharanthus roseus* [64], *Tagetes patula* [67], *Amaranthus cruentus* [68], *A. hypochondriacus* cvs. Rajagara and Rajagari [69], *A. hybridus* [70], lemongrass [71], *Marjorana hortensis* [72], *Ocimum basilicum* [73] and *Matthiola incana* [74].

Concerning the interaction between the effects of nitrogenous nutrition and different soils, data in Table 5 showed that, in both seasons, leaves of *Senna occidentalis* plants grown in Al-Gabal Al-Asfar soils and received Bio+N2 gave the highest values of N% (1.86 and 1.67% in the first and second seasons, respectively), P% (0.18 and 0.15%, respectively), K % (2.02 and 1.77%, respectively). In the second season, plants grown in Al-Gabal Al-Asfar soils and received Bio+N2 gave the same P% of that grown in Abou -Rawash soil and received Bio+N1. In both seasons, the leaves of plants grown in El-Tebbin soil without fertilization gave the lowest N% (1.04 and 1.15% in the first and second seasons, respectively), P% (0.05 and 0.06%, respectively) and K% (0.95 and 1.10%, respectively). In the second season, plants grown in El-Tebbin soil without fertilization gave the same P% that recorded with plants grown in Abou -Rawash soil and received biofertilization only.

Table 5: Effect of different soils as well as bio- and chemical N fertilizers on N, P and K (% D.W. of leaves) *Senna occidentalis*, Link. plants during 2010 and 2011 seasons

Fertilization treatments (F)	First season (2010)					Second season (2011)				
	Soils (S)					Soils (S)				
	S1	S2	S3	S4	Mean	S1	S2	S3	S4	Mean
	N (% D.W. of leaves)									
Control	1.28	1.31	1.11	1.04	1.19	1.22	1.29	1.19	1.15	1.21
N1	1.55	1.67	1.38	1.29	1.47	1.35	1.40	1.41	1.35	1.38
N2	1.75	1.78	1.46	1.36	1.59	1.49	1.58	1.56	1.43	1.52
Bio	1.37	1.45	1.25	1.09	1.29	1.29	1.39	1.11	1.22	1.25
Bio + N1	1.69	1.69	1.42	1.34	1.54	1.39	1.42	1.66	1.38	1.46
Bio + N2	1.79	1.86	1.59	1.38	1.66	1.40	1.67	1.49	1.45	1.50
Bio + 0.5 N1	1.45	1.56	1.28	1.16	1.36	1.30	1.38	1.26	1.23	1.29
Mean	1.55	1.62	1.36	1.24	---	1.35	1.45	1.38	1.32	---
	P (% D.W. of leaves)									
Control	0.09	0.09	0.06	0.05	0.07	0.08	0.09	0.07	0.06	0.08
N1	0.13	0.15	0.10	0.09	0.12	0.10	0.11	0.11	0.10	0.11
N2	0.16	0.16	0.11	0.10	0.13	0.12	0.13	0.13	0.11	0.12
Bio	0.10	0.11	0.08	0.06	0.09	0.09	0.10	0.06	0.08	0.08
Bio + N1	0.15	0.15	0.11	0.10	0.13	0.10	0.11	0.15	0.10	0.12
Bio + N2	0.17	0.18	0.13	0.10	0.15	0.11	0.15	0.12	0.11	0.12
Bio + 0.5 N1	0.11	0.13	0.09	0.07	0.10	0.09	0.10	0.08	0.08	0.09
Mean	0.13	0.14	0.10	0.08	---	0.10	0.11	0.10	0.09	---
	K (% D.W. of leaves)									
Control	1.26	1.30	1.04	0.95	1.14	1.19	1.28	1.15	1.10	1.18
N1	1.62	1.77	1.39	1.28	1.52	1.36	1.42	1.43	1.36	1.39
N2	1.88	1.91	1.50	1.37	1.67	1.54	1.65	1.63	1.46	1.57
Bio	1.38	1.49	1.23	1.02	1.28	1.28	1.41	1.04	1.19	1.23
Bio + N1	1.80	1.80	1.45	1.34	1.60	1.41	1.45	1.76	1.39	1.50
Bio + N2	1.93	2.02	1.67	1.39	1.75	1.42	1.77	1.54	1.49	1.56
Bio + 0.5 N1	1.49	1.63	1.26	1.11	1.37	1.29	1.39	1.24	1.20	1.28
Mean	1.62	1.70	1.36	1.21	---	1.36	1.48	1.40	1.31	---

S1: Fac. Agric., Giza.

S2: Al – Gabal Al- Asfar.

S3: Abou -Rawash.

S4: El-Tebbin

N1 = 2 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 4 g /pot till the end of the experiment

N2 = 4 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 8 g /pot till the end of the experiment

Bio= "Cerealin" containing *Bacillus polymyxa* and *Azotobacter chroococcum* bacteria.

Dry Weight of the above Ground Parts, Their Ni and Zn Concentration and Ni and Zn Uptake: The effect of different soils and fertilization treatments on dry weight of above ground parts (shoots and leaves) of *Senna occidentalis*, Link plants, Ni and Zn concentrations in the dry matter of above ground part tissues and their uptake are presented in Table 6. In both seasons, the dry weights of above ground parts of plants grown in most of the polluted soils (viz., Abou Rawash and El-Tebbin soils) were generally lower than that of plants grown in the un-polluted soil (Faculty of Agriculture soil). The only exception to this trend was recorded in both seasons with plants grown in the polluted soil of Al-Gabal Al-Asfar, which gave the heaviest dry weight of above ground parts, compared to plants grown in the other soils (in most cases). In each soil, in both seasons, raising the applied

nitrogen level caused a steady increase in the dry weight of above ground parts, with the highest values being found in plants received Bio+N2 treatment.

In both seasons, data presented in Table 6 revealed that the concentrations of Ni and Zn in dried above ground parts varied depending on the soil used. Generally, plants grown in polluted soils had higher Ni and Zn concentrations than the un-polluted soil. In most cases, under the same fertilization treatment, plants grown in El-Tebbin soil gave the highest Ni and Zn concentration followed by those grown in Abou Rawash and Al-Gabal Al-Asfar soils, whereas plants grown in Faculty of Agriculture soil had the lowest Ni and Zn concentrations. In general, the increase in Ni and Zn concentrations in dry matter of above ground parts of *Senna occidentalis* plants were associated with their

Table 6: Effect of different soils as well as bio- and chemical N fertilizers on dry weight of above ground parts of *Senna occidentalis*, Link. plants, their Ni and Zn concentration and uptake during 2010 and 2011 seasons

Soils	Fertilization treatments	First season (2010)					Second season (2011)				
		D.W (g/pot)	Ni		Zn		D.W (g/pot)	Ni		Zn	
			Conc. (µg/ g D.W)	Uptake ((µg/ pot)	Conc. (µg/ g D.W)	Uptake ((µg/ pot)		Conc. (µg/ g D.W)	Uptake ((µg/ pot)	Conc. (µg/ g D.W)	Uptake ((µg/ pot)
S1	Control	38.6	20	772	11.1	428.5	39.7	10	397	18.2	722.5
	N1	60.9	14	852	16.3	992.7	59.6	13	775	26.1	1555.6
	N2	74.0	13	962	29.0	2146.0	74.2	12	890	28.0	2077.6
	Bio	46.6	17	792	15.4	717.6	44.4	11	488	16.4	728.2
	Bio + N1	68.4	13	889	20.4	1395.4	65.6	13	853	31.0	2033.6
	Bio + N2	80.7	12	968	26.8	2162.8	79.0	12	948	36.0	2844.0
	Bio + 0.5 N1	50.7	16	811	19.0	963.3	51.9	11	571	22.8	1183.3
S2	Control	44.2	615	27183	31.2	1379.0	48.0	640	30720	19.7	945.6
	N1	62.0	630	39060	32.0	1984.0	67.9	679	46104	30.5	2071.0
	N2	80.2	680	54536	40.0	3208.0	85.3	650	55445	33.5	2857.6
	Bio	48.3	635	30671	29.4	1420.0	55.1	645	35540	25.8	1421.6
	Bio + N1	73.5	690	50715	36.0	2646.0	77.4	694	53716	35.7	2763.2
	Bio + N2	83.4	700	58380	32.8	2735.5	89.0	640	56960	39.8	3542.2
	Bio + 0.5 N1	56.0	595	33320	35.0	1960.0	61.0	610	37210	32.5	1982.5
S3	Control	29.0	754	21866	35.1	1017.9	32.1	820	26322	24.2	776.8
	N1	46.9	846	39677	38.0	1782.2	46.0	894	41124	36.7	1688.2
	N2	58.1	934	54265	55.0	3195.5	56.4	837	47207	46.7	2633.9
	Bio	35.3	730	25769	39.2	1383.8	35.0	790	27650	23.8	833.0
	Bio + N1	52.7	830	43741	40.0	2108.0	52.1	896	46682	41.2	2146.5
	Bio + N2	64.6	910	58786	50.8	3281.7	60.0	920	55200	56.8	3408.0
	Bio + 0.5 N1	38.7	700	27090	45.4	1757.0	37.6	750	28200	31.7	1191.9
S4	Control	26.1	940	24534	41.2	1075.3	26.0	837	21762	31.8	826.8
	N1	36.2	1167	42245	48.0	1737.6	36.9	850	31365	58.7	2166.0
	N2	46.5	1260	58590	52.0	2418.0	45.4	1260	57204	63.7	2892.0
	Bio	29.4	854	25108	39.4	1158.4	30.4	810	24624	43.8	1331.5
	Bio + N1	43.2	1305	56376	52.5	2268.0	41.2	1168	48122	53.1	2187.7
	Bio + N2	52.6	1200	63120	47.8	2514.3	48.3	1191	57525	68.9	3327.9
	Bio + 0.5 N1	32.8	924	30307	36.7	1203.8	32.3	821	26518	49.2	1589.2

S1: Fac. Ag. Giza, S2: Giza, S3: Kafrou -Rawash, S4: El-Tebbin

N1 = 2 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 4 g /pot till the end of the experiment

N2 = 4 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 8 g /pot till the end of the experiment

Bio= "Cerealin" containing *Bacillus polymyxa* and *Azotobacter chroococcum* bacteria

concentrations in the soil. Such results are in agreement with that reported on *Pelargonium zonale*, *Gazania splendens* and *Vinca rosea* [54], *Brassaia arboricola* var. *variegata* and *Ficus microcarpa* var. *Hawaii* [55], *Casuarina glauca*, *Taxodium distichum* and *Populus nigra* [65], *Quercus rotundifolia* [75], poplar [76] as well as *Amaranthus hybridus* and *A. dubius* [77].

Data presented in Table 6 revealed that the Ni concentrations in dried above ground parts of plants grown in polluted soils ranged from 595 to 1305 µg/g dry matter in the first season and from 610 to 1260 µg/g dry matter in the second season. These values are much higher than the values of 1000 µg/g dry weight reported for Ni hyperaccumulator [78]. The Ni concentrations in dried above ground parts of plants grown in polluted soils

were 49 to 76 times in the first season and 61 to 96 times in the second season higher than that of plants grown in non-polluted soil. On the other hand, the Zn concentrations in the dried above ground parts of plants grown in polluted soils were low and still in the normal range; they ranged from 29.4 to 55.0 µg/g dry matter in the first season and from 19.7 to 63.7 µg/g dry matter in the second season. These values were 1.9 to 2.6 times in the first season and 1.2 to 1.8 times in the second season higher than that of plants grown in non-polluted soil. Similar results were reported on *Betula papyrifera* and *Acer rubrum* [79] and salix [80]. Generally, no clear trend was noted for the Ni and Zn concentrations in the dried above ground parts of plants in response to N fertilization treatments in both seasons.

Table 7: DTPA extractable Ni and Zn ($\mu\text{g/g}$ soil) of the soils used in the experiment before and after treatments during 2010 and 2011 seasons

Soils	Fertilization treatments	DTPA extractable Nickel			DTPA extractable Zinc		
		Before	After		Before	After	
			2010	2011		2010	2011
S1	Control	0.2	0.05	0.12	7.0	6.9	6.8
	N1		0.03	0.05		6.8	6.5
	N2		0.01	0.02		6.6	6.4
	Bio		0.04	0.1		6.9	6.8
	Bio + N1		0.02	0.03		6.7	6.9
	Bio + N2		0.01	0.01		6.5	6.4
	Bio + 0.5 N1		0.04	0.09		6.8	6.8
S2	Control	12.1	6.6	5.8	91.0	90.7	90.8
	N1		4.3	2.7		90.6	89.8
	N2		1.2	1.1		89.3	89.4
	Bio		5.9	4.9		90.7	90.7
	Bio + N1		1.9	1.3		88.4	88.4
	Bio + N2		0.4	0.7		87.4	90.3
	Bio + 0.5 N1		5.4	4.6		90.6	90.6
S3	Control	12.3	7.9	7.1	95.0	94.7	92.8
	N1		4.4	4.1		94.0	94.7
	N2		1.4	2.9		93.6	90.5
	Bio		7.1	6.7		94.7	94.8
	Bio + N1		3.6	2.8		92.6	92.6
	Bio + N2		0.5	1.3		91.3	90.3
	Bio + 0.5 N1		6.9	6.5		93.0	94.8
S4	Control	13.2	8.3	8.6	99.0	97.7	97.8
	N1		4.7	6.7		94.6	96.6
	N2		1.5	1.7		93.5	95.4
	Bio		8.2	8.1		95.8	97.7
	Bio + N1		1.9	3.5		93.5	94.6
	Bio + N2		0.6	1.6		91.8	93.3
	Bio + 0.5 N1		7.1	7.9		97.7	97.7

S1: Fac. Agric., Giza. S2: Al – Gabal Al- Asfar. S3: Abou -Rawash. S4: El-Tebbin

N1 = 2 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 4 g /pot till the end of the experiment

N2 = 4 g ammonium sulphate (20.5 % N) /pot/month for 2 months and 8 g /pot till the end of the experiment

Bio= “Cerealin” containing *Bacillus polymyxa* and *Azotobacter chroococcum* bacteria

Data presented in Table 6 showed that in both seasons, the Ni and Zn uptake by *Senna occidentalis* plants was increased as a result of the different N nutrition treatments, because of the increase in dry weight of above ground parts of plants, compared to the control. Also, raising the applied nitrogen level caused a steady increase in the Ni and Zn uptake in most cases. Also, within each soil, plants received Bio+N1 or Bio+N2 treatments resulted in an increase in Ni and Zn uptake than plants received the same treatment without biofertilizer, in most cases. So, in general, plants received Bio+N2 treatment showed the highest Ni and Zn uptake, in both seasons. The results showed that plants extracted enormous amounts of Ni. Moreover, Bio+N2 treatment proved to be more efficient than the other treatments in remediating Ni polluted soils (i.e., taking up higher Ni

concentrations from the soil). On the other hand, the results showed that the plants extracted normal amounts of Zn. This means that *Senna occidentalis*, Link plants are non-hyperaccumulator plants for Zn and, therefore, cannot be used to remediate Zn polluted soils.

IV- DTPA Extractable Ni and Zn: The effect of different soils and fertilization treatments on extractable Ni and Zn ($\mu\text{g/g}$ soil) of the soils used in the experiment before and after treatments are presented in Table 7. In both seasons the extractable Ni and Zn of each tested soil used in the experiment after treatments were markedly reduced as a result of the different fertilization treatments, which increased the dry matter of *Senna occidentalis*, Link plants and their Ni and Zn uptake in most cases, compared to the unfertilized control. Also, in most cases,

raising the applied nitrogen level caused a steady decrease in the extractable Ni and Zn of the used soils, with the lowest values being found in plants supplied with Bio+N₂ treatment.

In most cases, El-Tebbin soil gave the highest extractable Ni and Zn after treatments followed by Abou Rawash and Al-Gabal Al-Asfar soils, whereas Faculty of Agriculture soil gave the lowest extractable Ni and Zn. Generally *Senna occidentalis*, Link plants reduced extractable Ni by 36% to 97% in the first season and 35 to 95% in the second season in the polluted soils. In contrast, *Senna occidentalis* plants caused only a negligible reduction in the extractable Zn in polluted soils (by 1 to 7% in the first season and 1 to 5.0% in the second one).

CONCLUSION

From the above results, it is clear that the best vegetative growth of *Senna occidentalis*, Link plants was obtained when the plants were grown in Al-Gabal Al-Asfar soil and received Bio+N₂ treatment. This treatment combination gave the highest values for most of the vegetative growth characteristics and chemical constituents. Generally, Bio+N₁ gave vegetative growth insignificantly different than that the best recorded with plants received Bio+N₂. It is also worth mentioning that, in each of the tested soils; applied Bio+N₂ gave the best results for most of the studied vegetative growth parameters, as well as for most of the plant chemical components and increased the ability of plants to remediate Ni polluted soils. However, the results showed that *Senna occidentalis*, Link plants cannot remediate Zn polluted soils.

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