# SEWAGE SLUDGE LEVELS ON THE DEVELOPMENT AND NUTRITION OF SUNFLOWER PLANTS

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### ABSTRACT

This study aimed to evaluate the effect of substituting chemical nitrogen (N) fertilization for equivalent N levels from sewage sludge of Wastewater Treatment Plant (WTP) on sunflower plant development. Nutrient levels in physiologically mature leaves and seeds, besides nutrient exportation during a 130-day assay, were also assessed. The experiment was carried out in 100 m<sup>2</sup> permanent plots at São Manuel Farm, which belongs to School of Agronomical Sciences, São Paulo State University-UNESP, Botucatu, São Paulo State, Brazil. The farm is located in the municipality of São Manuel, São Paulo State, Experimental design was in randomized blocks including 5 treatments and 5 replicates. Treatments were: T1 – chemical N fertilization according to the recommendation for the culture; T2 - 50% N from sewage sludge and 50% N from chemical fertilization; T3 - 100% N from sewage sludge; T4 - 150% N from sewage sludge; T5 - 200% N from sewage sludge. For all treatments, equal amounts of P and K fertilization were applied. Treatments differed for plant height from 21 to 64 days, stem diameter from 28 to 57 days, and leaf number from 21 to 38 days. Seed nutrient levels slightly varied; however, the quantities of exported N, P, Mg, Fe and Zn varied as sewage sludge levels increased.

Keywords: manure, biosolids and nitrogen

### INTRODUCTION

Sewage sludge is a residue from biological wastewater treatment plants, where domestic waste predominates over industrial one and heavy metal levels and pathogens are within the acceptable range for agricultural use (CONAMA, 2006). However, the final disposal of sewage sludge is a worldwide concern since its produced volume is increasing.

As an alternative, this residue has already been used for agriculture in numerous countries. The collection and treatment system for domestic waste is one of the basic assumptions for a healthy environment, providing life quality and environment preservation (Andreoli *et al.*, 1997).

Sewage sludge application was studied by Prado and Natale (2005) in passionflower culture. In a greenhouse, plants were subjected to sewage sludge levels of up to 30 mg ha<sup>-1</sup> (dry matter basis) and yielded quadratic responses for dry matter production and nutrient accumulation. In another study, a 4-fold N demand increased dry phytomass production and N level in *Brachiaria decumbens* leaves (Araújo *et al.*, 2009). In bean plants, N levels increased as the added sewage sludge levels were higher (Barros *et al.*, 2002).

Since sewage sludge is rich in nutrients, mainly nitrogen (N) and phosphorous (P), it has been used in many countries as a substitute for chemical fertilizers (Binder et al., 2002). Mineral nutrition is an important environmental factor and N is the macronutrient mostly required by cultures since plant growth and development are highly dependent on the availability of this nutrient. Such high dependence is due to the roles of N in plant metabolism, being a constituent of chlorophyll molecules, nucleic acids, amino acids and proteins (Taiz and Zeiger, 2004; Malavolta, 2006; Epstein and Bloom, 2006).

Lobo and Grassi Filho (2007) reported that in sunflower culture N from mineral fertilization can be substituted for that from sewage sludge, significantly increasing grain productivity, oil and dry matter yield.

Thus, the present work aimed to evaluate the effect of substituting chemical nitrogen fertilization for equivalent levels of N from WTP sewage sludge on sunflower development parameters, as well as to assess nutrient levels in physiologically mature leaves and seeds, including nutrient exportation.

# MATERIAL AND METHODS

This study was carried out at São Manuel Experimental Farm, which belongs to School of Agronomical Sciences, São Paulo State University–UNESP, Botucatu, São Paulo State, Brazil. The farm is located in São Manuel Municipality, São Paulo State (22° 25' S; 48° 34'W), at an altitude of 750 m above

According sea level. to Köppen classification, the climate in the study region is mesothermal. Cwa. i.e. humid subtropical including drought in the winter and rain from November to April. and the average annual rainfall in that municipality is 1,433 mm. The air relative humidity is 71%, with annual average temperature of 23°C. Meteorological data and classification were supplied by the Department of Natural Resources. Environmental Sciences Area, College of Agronomical Sciences-FCA, UNESP, Botucatu. The soil physical and chemical characteristics were already described by Lobo and Grassi Filho (2007).

Prior to the experiment stablishment, chemical analyses were done for the soil in the depth ranges of 0-20 to 20-40 cm, according to Raij *et al.* (2001). These results are shown in Tables 1 and 2.

 $P_2O_5$  and  $K_2O$  fertilizations were done at planting according to the analysis of soil with 30 kg ha<sub>-1</sub>  $P_2O_5$  in the form of simple superphosphate and 30 kg ha<sub>-1</sub>  $K_2O$  in the form of potassium chloride.

Experimental design was in randomized blocks constituted of 5 treatments and 5 replicates, as defined by Pimentel Gomes (2000): T1, chemical nitrogen fertilization of 50 kg ha-1, 10 kg being applied at planting and 40 kg N in cover fertilization, which is recommended for sunflower culture in São Paulo State, Brazil – 100% chemical nitrogen (urea); T2, 50% sewage-sludge nitrogen and 50% chemical nitrogen, cover fertilization (urea); T3, 100% sewage-sludge nitrogen fertilization, as recommended for the culture; T4, 150% sewage-sludge nitrogen fertilization; T5, 200% sewage-sludge nitrogen fertilization.

The used sewage sludge was from the Sewage Treatment Plant in the city of Jundiaí, São Paulo State. It was collected from samples composed of sludge presenting the characteristics shown in Table 3 (LANARV, 1988). The analysis was carried out in the Laboratory of Fertilizers and Correctors, Department of Natural Resources / Soil Sciences, School of Agronomical Sciences, Botucatu, São Paulo State.

Plot size, spacing, sewage-sludge N calculation, cultivar, sewage sludge chemical characteristics, and the remaining culture procedures were previously described by Lobo and Grasi Filho (2007).

Total rainfall during the crop season was 507 mm, reaching 296 mm in the first month of sowing.

Plant development parameters (plant height, leaf number and stem diameter) were weekly evaluated until the flowering phase. Plant height, stem diameter, leaf number, and macro and micronutrient levels in the leaves were analyzed together for 10 plants randomly distributed in each plot.

**Table 1.** Chemical characteristics of the soil where the experiment was developed (basic).

Depth	pН	O.M.	P(res.)	H+A1	Al <sup>3+</sup>	$K^+$	Ca <sup>2+</sup>	Mg <sup>2+</sup>	SB	CEC	BS
cm	CaCl <sub>2</sub>	g dm <sup>-3</sup>	mg dm <sup>-3</sup>			mi	mol <sub>c</sub> dm	.3			%
0-20	6.1	12	20	13	1	1.9	19	12	33	46	71
20 - 40	6.1	7	6	13	1	1.6	17	9	28	41	68

CEC= Ca+Mg+K+H+Al; SB= Ca+Mg+K; BS= ((SB/CEC)\*100). Raij et al. (2001).

**Table 2.** Chemical characteristics of the soil where the experiment was developed (micronutrients).

Depth	В	Cu	Fe	Mn	Zn					
cm		mg dm <sup>-3</sup>								
0-20	0.11	1.0	20	7.7	1.2					
20-40	0.09	0.9	13	4.1	1.2					

Table 3. Chemical characteristics of the sewage sludge used in the experiment.

Ν	$P_2O_5$	$K_2O$	Humidity.	MO	С	Са	Mg	S	Na	Cu	Fe	Mn	Zn	C/N	pН
			g kg <sup>-1</sup>								- mg kg	-1			
-															
31.8	17.2	1.8	675.8	550	306	12.5	2.2	45.6	1520	812	31650	3400	2150	10/1	4.3

Plant height was first measured at 20 d after sowing and weekly repeated until 70 days by using a tape measure of 100 cm maximal height; from this maximal point a 2 m ruler was employed. During the vegetative development, height was assessed from the soil to the insertion of the last unfolded leaf, while in the reproductive phase it was measured from the soil to the capitulum insertion. Stem

diameter (cm) was first measured at 34 days after sowing and weekly repeated until 70 days by using a caliper from the soil. Leaf number was also weekly determined from 27 to 56 d after sowing by counting the number of completely unfolded leaves in the plant.

For macro and micronutrient analysis leaves were harvested at the beginning of flowering (Malavolta *et al.*, 1997), and the 4<sup>th</sup> leaf from the upper part of the plant was collected. Harvested leaves were analyzed for N, P, K, Ca, Mg, S, B, Cu, Fe, Mn and Zn levels, according to the methodology described by Malavolta *et al.* (1997).

Seeds were collected from 10 plants in the stage R9, when capitula were turned down. They were dried in a forced aeration oven at  $60^{\circ}C \pm 5^{\circ}C$  for 7 d and ground by using a Willey-type mill. The ground material was stored in paper bags and sent for macro and micronutrient analysis at the Laboratory of Plant Mineral Nutrition, Department of Natural Resources/Soil Sciences, FCA/UNESP, where N, P, K, Ca, Mg, S, B, Cu, Fe, Mn and Zn levels were determined according to the methodology of Malavolta et al. (1997). Results were used to calculate the levels of nutrients exported by the seeds as follows: the level of each element was multiplied by each plot productivity in 1 ha

Data were subjected to comparison of means by Tukey's test at 5% probability level, according to Statistical Analysis System procedures (SAS Institute, 2001).

## **RESULTS AND DISCUSSION**

Mean plant heights on emergence days 14, 21, 28, 38, 44, 50, 57 and 64 are represented in Figure 1. From 21 to 38 d, T4 and T5 had superior heights, compared to the other treatments. From 38 to 50 days, T5 presented the highest height. From 50 d after emergence, mean heights were higher in T5 and T2. In any moment, T5 was inferior to the other treatments, providing better land closing and not allowing weeds; in addition, the increased height development could protect the soil for a longer period. Salvador (2006) used sludge containing 0.63% N and 66.73% dry matter and at 83 days after planting the height of maize plants receiving 40 mg ha<sup>-1</sup> sewage sludge on a wet weight basis was equal to that of plants receiving complete mineral fertilization.

Similarly, Tanaka (1981) reported a beneficial effect of N on sunflower culture, increasing plant height. Biscaro *et al.* (2008) obtained a quadratic response of N application for height at 30 d after emergence; the application of 69 kg ha<sup>-1</sup> N led to a 40.6 cm height. At 45 d after emergence, 114.7 cm height was obtained with the application of 72.9 kg ha<sup>-1</sup> N. In the latter, cover nutrient application was important to plant growth and resulted in a good plant size, without lodging and with easy management and harvest due to the appropriate height.

As shown in Figure 2, at 28 d after emergence stem diameter was significantly greater in T5 (1.4 cm) than in T1 (1.1 cm), T2 (1.1 cm) and T3 (1.2 cm). At 38 and 44 d, T5 had the greatest diameter (2.0 and 2.4 cm, respectively). At 50 and 57 d, respectively, diameter was statistically equal in T2 (2.4 and 2.6 cm) and T5 (2.7 and 2.8 cm), which was probably due to the nitrogen fertilization in T2; besides, T5 diameters were statistically greater than those of T1 (2.4 and 2.4 cm), T3 (2.2 and 2.4 cm) and T4 (2.3 and 2.5 cm).

Biscaro *et al.* (2008) also applied N up to the estimated maximal level of 47.8 kg ha<sup>-1</sup> in sunflower plants and obtained increased stem diameters, which reached a mean growth of 18.4 mm. Stem diameter is an important characteristic for sunflower plants since it allows less lodging, facilitating management and harvest.

As regards leaf number (Figure 3), already in the first evaluation, at 21 d after emergence, T2, T3, T4 and T5 had a mean leaf number of 12.8, 12.6, 12.8 and 13.8, respectively, and did not differ statistically; T1 had a mean number of 11,



Figure 1. Mean height of sunflower plants expressed as days after emergence



Figure 2. Mean stem diameter of sunflower plant from 28 to 57 days after emergence



Figure 3. Mean leaf number of sunflower plants from 21 to 50 days after emergence

which was statistically smaller than that of the other treatments. At 28 d after emergence, T5 had 21.1 leaves per plant and did not differ from T4, which had 20.2 leaves per plant; in addition, the mean leaf number of T5 was larger than that of T1, T2 and T3, which presented 17.8, 18.8 and 19.3 leaves per plant, respectively, and leaf number in T3 was superior to that in T1. At 44 and 50 d, all statistically treatments were equal. presenting a mean leaf number of 30.1 and 32, respectively; these latter values probably did not vary due to the N fertilization in T1 and T2.

According to Biscaro *et al.* (2008), there is a significant effect of cover N application in sunflower. At 30 d after emergence, those authors obtained a quadratic response of the applied N levels with the highest level of 80 kg ha-1, and leaf number increased up to 15.5 leaves per plant. A significant effect was also verified at 45 d after emergence, including a quadratic response of N level with the highest studied level (80 kg ha<sup>-1</sup>), resulting in 29.2 leaves per plant. This indicates that the leaf number per plant was influenced by cover N application, following plant growth and producing leaves until the plant reached its maximal growth.

Primary macronutrients (N, P and K) in the leaves did not statistically differ among treatments. N levels were adequate  $(35-50 \text{ g kg}^{-1})$ , and the mean among treatments was 34 g kg<sup>-1</sup>, indicating no problem due to the lack of N in the leaf harvest period. P mean among treatments was 5 g kg<sup>-1</sup> and did not vary; however, the adequate level is from 2.9 to 4.5 g kg<sup>-1</sup> (Castro and Oliveira, 2005). K mean among treatments was 40.3 g kg<sup>-1</sup> and all treatments had adequate levels  $(31 - 45 \text{ g kg}^{-1})$  (Castro and Oliveira, 2005), as shown in Table 4.

Treatment	Ν	Р	K	Ca	Mg	S				
g kg <sup>-1</sup>										
T1	35.8 a	4.6 a	41.2 a	19.0 a	5.9 a	3.2 a				
T2	36.0 a	5.1 a	40.2 a	21.6 a	6.8 a	3.6 a				
Т3	32.8 a	5.2 a	42.6 a	20.4 a	6.5 a	3.7 a				
T4	33.4 a	5.0 a	40.6 a	20.6 a	7.0 a	3.9 a				
Т5	32.2 a	4.9a	36.8 a	20.4 a	6.3 a	3.8 a				
F	1.99 <sup>NS</sup>	0.88 <sup>NS</sup>	2.53 <sup>NS</sup>	0.45 <sup>NS</sup>	2.09 <sup>NS</sup>	2.34 <sup>NS</sup>				
Mean	34.0	5.0	40.3	20.4	6.5	3.6				
CV	8.1	11.8	7.5	15.1	10.1	10.7				

Table 4. Mean levels of N, P, K, Ca, Mg and S in sunflower leaves

Equal lowercase letters in the columns do not differ according to the Tukey's test at 5% probability level. NS: Non-Significant

Secondary macronutrients (Ca, Mg and S) did not differ among treatments and were present at adequate levels: Ca, 19 to 32 g kg<sup>-1</sup>; Mg, 5.1 to 9.4 g kg<sup>-1</sup>; and S, 3 to 6.4 g kg<sup>-1</sup> concerning leaf levels for sunflower culture (Castro and Oliveira, 2005). The vaules of Ca, Mg and S were adequate: 20.4, 6.5 and 3.6, respectively.

According to Castro and Oliveira (2005),the adequate levels for micronutrients are: B, 35-80 mg kg<sup>-1</sup>; Cu, 24-42 mg kg<sup>-1</sup>; Fe, 120-235 mg kg<sup>-1</sup>; Mn, 55-180 mg kg<sup>-1</sup>; and Zn, 29-43 mg kg<sup>-1</sup>. B levels did not vary among treatments and were adequate (69.76 mg kg<sup>-1</sup>). Although Cu levels did not vary among treatments, they were slightly low (22.72 mg kg<sup>-1</sup>). Fe and Mn levels did not vary among treatments and were adequate (134.84 mg kg<sup>-1</sup> and 143.76 mg kg<sup>-1</sup>, respectively). Zn was the only element differed with that sewage sludge application since the latter contained a high level of this nutrient and the soil already had 1.2 mg dm-3 Zn, which is considered high (Raij et al., 1997). There was a significant difference between the treatment that did not receive sewage sludge and the ones that received it. The lowest sewage sludge level contained 5.2 kg ha<sup>-1</sup> Zn, and all leaf levels had high concentration of this element. The high Zn level in the soil and in the sewage sludge probably inhibited Cu absorption; thus, in the leaves Zn levels were high and Cu levels low (Table 5).

As shown in Table 6, macronutrient levels in the seeds did not vary. However, macronutrient exportation significantly differed as productivity increased, except for Ca which did not vary among treatments. This can be explained by the original high level of Ca in the soil, when the needed Ca level for sunflower to complete its cycle is too small, 9 kg ha<sup>-1</sup> (Castro and Oliveira, 2005).

As regards micronutrients, the only element that showed significant differences among treatments was Zn (Table 8). Micronutrient exportation by the seeds (Table 9) significantly increased as the applied sewage sludge levels were higher, except for Cu. Vieira *et al.* (2005) noticed that in soybean plants, up to 6 ton ha<sup>-1</sup> sewage sludge on the dry matter basis did not increase Cu, Zn and Fe levels in the grains.

Treatment	В	Cu	Fe	Mn	Zn			
	mg kg <sup>-1</sup>							
T1	70.6 a	22.8 a	144.4 a	130.6 a	45.6 b			
T2	75.4 a	24.0 a	143.8 a	141.2 a	49.8 a			
Т3	60.2 a	22.0 a	129.2 a	156.8 a	47.2 a			
T4	68.0 a	22.4 a	132.4 a	147.0 a	50.4 a			
Т5	74.6 a	22.4 a	124.4 a	143.2 a	50.6 a			
F	2.21 <sup>NS</sup>	0.71 <sup>NS</sup>	2.25 <sup>NS</sup>	0.29 <sup>NS</sup>	1.98 *			
Mean	69.76	22.72	134.84	143.76	48.72			
CV	13.2	9.0	9.9	27.5	7.2			

Table 5. Mean levels of B, Cu, Fe, Mn and Zn in sunflower leaves

Equal lowercase letters in the columns do not differ according to the Tukey's test at 5% probability level. \*significant at 5% probability level. NS: Non-Significant.

Treatments	Ν	Р	K	Ca	Mg	S
			g	kg <sup>-1</sup>		
T1	26.8 a	7.6 a	12.6 a	2.4 a	4.3 a	1.6 a
T2	29.0 a	7.8 a	11.6 a	2.0 a	4.2 a	1.5 a
Т3	29.0 a	8.3 a	12.2 a	2.0 a	4.5 a	1.6 a
T4	28.0 a	8.0 a	12.2 a	2.0 a	4.4 a	1.6 a
Т5	32.4 a	9.1 a	12.6 a	2.2 a	4.9 a	1.7 a
F	0.56 <sup>NS</sup>	2.32 <sup>NS</sup>	0.67 <sup>NS</sup>	1.45 <sup>NS</sup>	1.79 <sup>NS</sup>	1.47 <sup>NS</sup>
Mean	29.0	8.16	12.24	2.2	4.4	1.6
CV	21.5	10.7	9.1	15.1	16.6	8.1

Table 6. Mean levels of N, P, K, Ca, Mg and S in sunflower seeds

Equal lowercase letters in the columns do not differ according to the Tukey's test at 5% probability level. NS: Non-Significant

Treatments	Ν	Р	K	Ca	Mg	S
				kg ha <sup>-1</sup>		
T1	100.1 b	28.5 c	47.4 a	9.1 a	16.2 b	6.1 a
T2	140.7 ab	37.9 b	56.3 a	9.7 a	20.6 ab	7.3 a
Т3	129.3 ab	36.5 b	54.4 a	9.0 a	19.9 ab	7.2 a
T4	122.7 ab	35.1 bc	53.5 a	8.8 a	19.1 ab	7.0 a
Т5	165.3 a	46.6 a	64.7 a	11.2 a	25.0 a	8.8 a
F	3.34 *	3.5 *	2.0 <sup>NS</sup>	1.22 <sup>NS</sup>	3.27 *	2.35 <sup>NS</sup>
Mean	125.0	80.1	63.7	9.3	19.1	6.9
CV	23.6	21.0	17.8	21.4	20.6	20.4

Table 7. Macronutrient exportation by sunflower seeds

Equal lowercase letters in the columns do not differ according to the Tukey's test at 5% probability level. \*Significant at 5% probability level. NS: Non-Significant

Treatments	В	Cu	Fe	Mn	Zn
			mg kg <sup>-1</sup>		
T1	32.8 a	17.2 a	65.0 a	32.4 a	66.2 b
T2	33.2 a	16.0 a	62.8 a	27.0 a	70.2 b
Т3	31.4 a	15.4 a	68.4 a	35.0 a	72.0 b
T4	32.0 a	19.8 a	68.0 a	33.6 a	75.4 ab
Т5	35.6 a	20.6 a	73.4 a	33.8 a	89.8 a
F	0.78 <sup>NS</sup>	0.79 <sup>NS</sup>	0.74 <sup>NS</sup>	1.04 <sup>NS</sup>	6.04 *
Mean	33.5	17.8	67.6	32.2	73.3
CV	12.1	27.6	15.4	21.3	11.2

Table 8. Mean levels of B, Cu, Fe, Mn and Zn in sunflower seeds

Equal lowercase letters in the columns do not differ according to the Tukey's test at 5% probability level. \*Significant at 5% probability level. NS: Non-Significant

Seed nutrient levels did not significantly vary with sewage sludge application, except for Zn (Tables 6 and 7); however, the exported values showed significant differences, except for K, Ca, S, B and Mn (Tables 8 and 9).

Nutrient exportation is extremely important to quantify nutrients that are removed from the culture and to replace them, if necessary, in a following planting through comparison with the remaining levels in the soil. nutrient Although demand bv sunflower culture was high. its exportation by the seeds is low, i.e. several nutrients return to the soil through leaves, stems, roots and capitula after harvest; thus. the subsequent cultures present high

Treatments	В	Cu	Fe	Mn	Zn
		g ha <sup>-1</sup>			
T1	124.7 a	65.2 bc	245.6 c	122.7 a	247.4 c
T2	161.0 a	77.5 b	307.3 b	130.9 a	340.8 b
Т3	140.0 a	58.8 c	310.2 b	153.3 a	321.9 b
T4	143.0 a	86.3 b	299.7 b	142.9 a	329.9 b
Т5	181.0 a	102.4 a	376.6 a	170.2 a	456.1 a
F	2.28 <sup>NS</sup>	3.21*	90.42*	1.85*	5.9*
Mean	143.8	78.0	192.3	136.7	317.7
CV	22.2	27.7	9.0	24.1	21.7

 Table 9. Micronutrient exportation by sunflower seeds

Equal lowercase letters in the columns do not differ according to the Tukey's test at 5% probability level. \*Significant at 5% probability level. NS: Non-Significant

productivity. Castro *et al.* (2005) reported that sunflower plants had reduced nutrient exportation rates and higher dry matter and nutrient levels in plant remnants.

### CONCLUSIONS

The application of 30.4 t ha<sup>-1</sup> of the swage sludge on wet weight basis t (T5) led to higher vegetative development (height, stem diameter and leaf number) and Zn levels both in seeds and leaves, compared to the other treatments.

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