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Sewage Sludge Application in Soil Improved Leafy Vegetable Growth

Maryam Haghighi*
Department of Horticulture, College of Agriculture, Isfahan University of Technology, Isfahan, IRAN

ABSTRACT

A greenhouse experiment was conducted to investigate the effect of sludge application on growth rate of three leafy vegetable spinach, celery and lettuce. Treatment design in control and 0.7 mg/kg dry weight of soil. Results showed that sludge was highly effective in celery growth compare to spinach and lettuce and increased dry and fresh weight of celery significantly. Photosynthesis rate improved after 4 and 8 weeks in all three vegetables. Antioxidant activity showed that positive effect of sludge in availability of nutrient and prevention the stress of nutrient shortage in plants.

Keywords: Lettuce (*Lactuca sativa*), celery (*Apium graveolens* var. dulce), spinach (*Spinacia oleracea*), Superoxide dismutase (SOD) activity, Peroxides activity

INTRODUCTION

The application of organic-rich waste material such as sludge to agricultural soils can have a beneficial effect on soil biological, chemical and physical properties, plant growing, soil conditioning. They are rich in nitrogen, phosphorus, and other plant nutrients like boron, manganese, copper, molybdenum and zinc depending on the specific nature of the sludge material (Dolgen et al. 2004). Therefore, there is an increasing interest in the agricultural use of treatment plant with sludge. On the other hand, application of industrial wastewater for agriculture purposes can increased heavy metals in polluted places. These facts have discouraged the application of industrial wastewater treatment plant sludge for agriculture. There are many studies have been done on the toxicity and heavy metal availability of sludge. Conversly, the deleterious effect of sludge application depends on the chemical form and concentration of the metal species of sludges. Zheljazkov and Warman (2004) showed that mature Municipal Solid Waste Compost with concentrations of Cu and Zn of 311 and 767 mg/kg respectively could be used as a soil conditioner without phytotoxic effects on agricultural crops and without increasing the normal range of Cu and Zn in Basil and swiss chard. Haghighi et al. (2010) showed that organic matter application in soil immobilized Cd and reduced its toxicity in lettuce. In this study sludge has low heavy metal concentration from New Zealand Palmerston North City Council to investigate its effect on vegetable growth. So the results of this research can be used for land application of sludge in agricultural purposes in clean industrial places.

MATERIALS AND METHODS

Lettuce (*Lactuca sativa*), celery (*Apium graveolens* var. dulce) and spinach (*Spinacia oleracea*) was planted in pot contained 0.7 kg soil. 12 months old sludge was collected from the Palmerston North City Council. Two Treatments were arranged; control and 0.7 mg/kg dry sludge in dry weigh of soil applied to pots. The experiment was conducted from May 2008 at the Plant Growth Unit, Massey University, Palmerston North (lat. 40.2 ^{o}S, long. 175.4 ^{o}E), New Zealand the concentration of two main heavy metal in sludge Zn and Cu come in table 1. The experiment was housed in a naturally-lit glasshouse with ventilation/heating set points of 25/15 ^{o}C.

Table 1. The concentration of Zn and Cu of soil with sludge

Treatments	Zn Conc. of soil at (μg/g dry weight of soil)	Cu Conc. of soil (μg/g dry weight of soil)
C	6.7327	32.30
Sludge	6.6668	34.007

Immediately after plant harvesting, Fresh weight was measured with analytical balance and put in the oven with 70 ^{o}C. After they dried, dry weight was weighted.

* Corresponding author: haghighi@shirazu.ac.ir

Plant grew for 8 weeks and superoxide dismutase and peroxidas activity was assayed, according to Haghighi et al. (2010). Sub samples of supernatant (0.5 g) were homogenized in 5 mL of extraction buffer consisting of 50 mM phosphate (pH 7.8), 0.1% (w/v) BSA, 0.1% (w/v) ascorbate, 0.05% (w/v) mercaptoethanol. The assay mixture in 3 mL contained 50 mM phosphate buffer (pH 7.8), 9.9 mM L-methionine, 57 µM NBT, 0.025% (w/v) Triton X-100, and 0.0044 (w/v) riboflavin. The photo-reduction of NBT (formation of purple formazan) was measured at 560 nm. One unit of SOD activity was defined as that being present in the volume of extracts that caused inhibition of the photo-reduction of NBT by 50%.

The reaction mixture for Peroxide activity consisted of 50 mM potassium phosphate buffer (pH 6.1), 1% guaiacol (w/v), 0.4% H_2O_2 (v/v) and enzyme extract. The absorbance increase due to oxidation of guaiacol at 470 nm was measured. Enzyme activity was calculated as µM of guaiacol oxidized min^{-1} (g fresh weight)$^{-1}$ at 25 ± 2°C.

Photosynthesis rate was measured using a portable gas exchange system (Li-6200, LiCor, Lincoln, NE) 4 (Pn1) and 8(Pn2) weeks after seed planting and report as µmol CO_2 m^{-2} s^{-1}.

Experiment was design in CRD for each plant with 4 replicate. All data were compared by T-test Comparisons with P values of <0.05 being considered significantly different for treatments.

RESULTS AND DISCUSSION

Results revealed that fresh shoot of celery and spinach and dry weight of celery shoot increased with application of sludge but there were not observed any significant effect on lettuce fresh and dry weigh, and dry weight of spinach (Table 2).

SOD activity of spinach and POD activity of lettuce decreased along with application of sludge. SOD and POD activity of celery, POD of spinach and SOD of lettuce did not changed with sludge application (Table 2). Photosynthesis rate increased in lettuce, spinach and celery exposed to sludge after 4 and 8 weeks (Table 3).

Table 2. The effect of sludge application on fresh and dry weight, POD and SOD activity of leafy vegetable

Treatments	Fresh shoot of celery (g)	Dry shoot of celery (g)	Fresh shoot of spinach (g)	POD activity (U g^{-1} W) lettuce	SOD activity (U g^{-1} W) spinach
C	1.30	0.19	2.22	6.243	362.272
Sludge	4.36	4.36	3.70	3.594	223.291
T-test	0.035	0.021	0.002	0.059	0.045
STDEV	1.884	0.218	0.848	1.545	12.655

Table 3. The effect of sludge application on photosynthesis rate (Pn) of leafy vegetable after 4 and 8 weeks

Treatments	lettuce		Spinach		celery	
	Pn1	Pn2	Pn1	Pn2	Pn1	Pn2
C	16	9.95	13.55	15.95	8.35	12.7
Sludge	22.1	17.75	13.15	29.7	17.3	18.5
T-test	0.017	0.022	0.949	0.042	0.047	0.046
STDEV	3.553	4.609	4.139	8.277	5.407	3.374

Sludge potentially improve growth as reported with other researcher; Compost addition improved the growth of ryegrass and tall fescue (*Festuca arundinacea*) (Park et al. 2011).Singh and Agrawal (2008) reviewed the beneficial effect of sludge on growth, fresh and dry weight and photosynthesis rate of some plants (sunflower, maize, barely etc.). Along with their results, in this study, the comparison control and treatment with sludge showed that sludge application increase fresh shoot weight due to improvement of photosynthesis rate of some of leafy vegetable and nutrient uptake. Although there are some reports that's reveald that sewage sludge did not affect *Beta vulgaris* the same results was observed for lettuce in this study. Generally organic matter increase plant micronutrient uptake like Fe, Zn, and Cu in low and essential concentration reported in cucumber, melon and bentgrass due to their ability to complex metals and promote plant growth (Garcia-Mina et al, 2004). To the best of our knowledge there are not any field research investigate the effect of clean sludge in the point of view of heavy metals on antioxidant activity of leafy

vegetables. Relevantly, the result of this study showed that shortage of nutrient availability and uptake in control treatment induced antioxidant activity in this treatment more. Pant et al. (2009) reported that pak choi treated with vermicompost has lower antioxidant activity than control. They suggested that low plant growth and N concentration caused high level of antioxidant activity of leafy vegetables. There are many different study illustrated the improvement effect of organic matter on photosynthesis rate. The improvement effect of sludge as a organic matter because of some reasons come to Nardi et al (2002) review on humic acid organic matter induced photosynthesis rate by enhancing chlorophyll content and interfere in enzymes activity involved in carbohydrate metabolism and later used in photosynthesis' cycle. Finally, organic matter increased photosynthesis rate by stimulation of enzyme activities related to the photosynthetic sulphate reduction pathway. Zheljazkov and Warman (2004) showed that mature Municipal Solid Waste Compost with concentrations of Cu and Zn of 311 and 767 mg/kg respectively could be used as a soil conditioner without phytotoxic effects on agricultural crops and without increasing the normal range of Cu and Zn in Basil and swiss chard relevantly in this study Cu and Zn concentration of soil amendment with sludge was 34.00 and 6.66 µg/g dry weight of soil, respectively (Table 1) and there were not observed any toxic effect on celery, spinach and lettuce.

CONCLUSIONS

Clean sludge from heavy metal can improve growth of celery more than lettuce and spinach; because celery growth time is longer than spinach and lettuce so it is exposed to media including sludge more effectively. Therefore, more quantity of sludge for spinach and lettuce can be more effective. The effect of sludge in growth rate is via improving photosynthesis rate of all three vegetables. Although the concentration of elements did not measured in this study but resulting from other research sludge through increasing element uptake induced photosynthesis and growth of leafy vegetable. Enhancing SOD and POD activity in control compare to sludge application improved this hypothesis that control treatment exposed to shortage of nutrient element compare to sludge treated plants.

REFERENCES

Dolgen D, Alpaslan M N, Delen N (2004). Use of an agro-industry treatment plant sludge on iceberg lettuce growth. Ecological Engineering. 23: 117–125.

Garcia-Mina JM, Antolin MC and Sanchez-Diaz M (2004). Metal-humic complexes and plant micronutrient uptake: a study based on different plant species cultivated in diverse soil types. Plant and Soil. 258: 57–68.

Haghighi M, Kafi M, Fang P, Gui-Xiao L (2010).Humic acid decreased hazardous of Cadmium toxicity on lettuce (*Lactuca sativa* L.). Vegetable Crops Res. Bulletin. 72: 49-61.

Nardi S, Pizzeghello D, Muscolo A, Vianello A (2002). Physiological effects of humic substances on higher plants Soil Biology & Biochemistry. 34 : 1527–1536.

Pant A P, Radovich T JK, Hue N V, Talcottb S T and Krenekb K A (2009). Vermicompost extracts influence growth, mineral nutrients, phytonutrients and antioxidant activity in pak choi (*Brassica rapa* cv. Bonsai, Chinensis group) grown under vermicompost and chemical fertilizer. J Sci Food Agric . 89: 2383–2392.

Park J, Lamb D, Paneerselvam P, Choppala G, Bolan N, Chung JW (2011). Role of organic amendments on enhanced bioremediation of heavy metal contaminated soils. Journal of Hazardous Materials. 185. 549–574.

Singh A, and Agrawal M (2010).Effects of municipal waste water irrigation on availability of heavy metals and morpho-physiological characteristics of Beta vulgaris L. Journal of Environmental Biology. 31(5) 727-736.

Singh RP, Agrawal M (2008). Potential benefits and risks of land application of sewage sludge. Waste Management. 28: 347–358

Zheljazkov VD, and Warman PR (2004). Source-Separated Municipal Solid Waste Compost Application to Swiss Chard and Basil. J. Environ. Qual. 33:542-552.