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Effects of Sewage Sludge on the Seed Emergence, Development and Mineral Contents of Pepper (*Capsicum annum*) Seedling

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Abstract: This study was carried out to determine the effects of sewage sludge on the seed germination, seedling development and macro-micro nutrient contents of pepper seedling. Seedling growing media consisted of 40:20:40 Soil (S), Pumice (PM) and organic fertiliser mixture [Farmyard Manure (FM)+Sewage Sludge (SS)]. No additional nutrient was applied to the treatments. It was determined that sewage sludge had a positive effect on the seed emergence percentage and pepper seedling growth. FM and SS ratio in the growth media mixture affected the seedling growth parameters and mineral contents of peppers. As increased the SS ratio in the growth media mixture, seedling growth parameter and macro and micro nutrient contents of peppers were increased. Farmyard manure and SS application at 25% FM+75% SS ratio in the growth media mixture were effective combination in seedling parameter, but 50% FM+50% SS ratio was suitable for agricultural process in terms of heavy metal contents of plant.

Key words: Heavy metal, pepper, seedling growth, sewage sludge

INTRODUCTION

Application of sewage sludge to agricultural land has be common practice over the past several decades. This practice is inexpensive, logical and easy to carry out. Since sewage sludge contains plant nutrients and organic matter, it may be used to supplement or replace commercial fertilisers for crop production. The beneficial effects of using sludge on agriculture have been proven by numerous researchers. It has been shown that sewage sludge application improves the physical, chemical and biological properties of soil[1-3].

The use of sludge in agriculture and for land reclamation is increasingly being identified as an important issue for both soil conversation and residual disposal. Most sewage wastes contain valuable nutrients that could be used to improve soil fertility. Furthermore beneficial changes may be seen in soils with low organic matter contents which are prone to soil structure degradation as well as loss of soil fertility[4-6].

Recycling of sewage sludge to agricultural land is generally considered to be the best practicable environmental option[7] and in general, it has been shown that the addition of sludge to agricultural land enhances growth and yield of crops. On the other hand, sewage sludge application may lead to the accumulation of a number of potentially harmful components such as heavy metals in soil and crops. The prince of heavy metals in the applied sludge can result in phytotoxic effects, soil and water contamination and accumulation of heavy metal in food supplies[8,9]. It is often argued that heavy metals such as cadmium, nickel or lead in sludge, when applied to soils, may enter the food chain through plants or animals, contaminate surface and ground water and thus cause health hazards[10]. In reality, metal concentrations in sewage sludge vary widely depending on several factors, including sludge origin and sludge pre-treatment processes and the solubility or bioavailability of heavy metals from sewage sludge is based on soil pH, lime content, soil cation change capacity and soil organic matter.

The objectives of this study were to (I) assess the suitability of sewage sludge to supply some essential plant nutrient, (ii) determine an optimum application rate of sewage sludge and farmyard manure (iii) evaluate and compare the effects of farmyard manure application to reduce the heavy metal solubility of sewage (iv) seed emergence, seedling growth parameter and chemical composition of pepper seedling.

MATERIALS AND METHODS

The experiment was conducted to determine effects of sewage sludge on the seed emergence, development,

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macro and micro nutrient contents of pepper seedling in 2000. The seedlings were kept were maintained in a heated greenhouse under natural light at a minimum temperature of 19-21°C and maximum of 25-32°C. Day length was 14 h during the experimental period.

The experiment was conducted with Ilıca-256 pepper cultivar and carried out in the mixtures of 40:20:40 Soil (S), Pumice (PM) and organic fertiliser mixture [Farmyard Manure (FM)+Sewage Sludge (SS)] in 15 cm diameter polyethylene pots. The mixtures used as seedling growth medium as follows:

- 40% soil (S)+20% pumice (PM)+40% organic fertiliser (100% FM+0% SS),
- 40% soil (S)+20% pumice (PM)+40% organic fertiliser (75% FM+25% SS),
- 40% soil (S)+20% pumice (PM)+40% organic fertiliser (50% FM+50% SS),
- 40% soil (S)+20% pumice (PM)+40% organic fertiliser (25% FM+75% SS),
- 40% soil (S)+20% pumice (PM)+40% organic fertiliser (0% FM+100% SS).

Sewage sludge was digested and dewatered sewage sludge obtained from Van Wastewater Treatment Plant (WTP) Eastern part of Turkey. The chemical and physical properties of sewage sludge and farmyard manure were given in Table 1.

Sewage sludge was sun dried for 10 days to reduce moisture content before application. The sludge was the applied and incorporated to the soil, pumice and FM combination before 5 weeks seed sowing. The experiment was continued for six weeks after seed sowing. Seed germination percentage and period, hypocotyls length, cotyledon length and width were determined at the first week. Seedling length, fresh and dry seedling weights, root length, fresh and dry root weights, stem diameter and leaf number were determined at the end of the sixth week. Randomized Block Design was applied with three replications and each replication had five pots. In order to determine the mineral contents of plant shoot and root, plants samples were oven-dried at 68°C for 48 h and then ground. K^+, Ca^{+2} and Mg^{+2} were determined after wet digestion of dried and ground sub-samples in a H_2SO_4-Se-salisilic acid mixture. In the diluted digests, P was measured spectrophotometrically by the indophenol-blue method and after reaction with ascorbic acid. Potassium and Ca^{+2} were determined by flame photometry, Mg^{+2}, Mn, Zn, B, Mo, Cd and Cu by atomic absorption spectrometry using the method of AOAC[11]. All data were subjected to

Table 1: Some properties of sewage sludge and farmyard manure

	Sludge				Farmyard manure			
Properties	Max	Min.	Mean	SD	Max.	Min.	Mean	SD
Organic Matter (g/100 DM)	25	24	24.43	0.51	36.5	32.5	34.25	1.84
pH	6.1	5.79	5.98	0.13	5.94	5.5	5.74	0.18
Total N (g/100 DM)	1.3	1.08	1.21	0.13	0.45	0.38	0.41	0.03
Available N (mg/kg DM)								
NH_4-N	1795	1650	1717.5	60.34	925	785	861.30	59.35
NO_3-N	254	235	246.5	8.18	925	785	861.25	59.35
Total P (g/100 DM)	1.59	1.49	1.53	0.05	0.17	0.12	0.14	0.02
Available P (mg/kg DM)	1061	900	984.75	66.52	420	366	387.5	23.08
Total K (g/100 DM)	0.41	0.38	0.39	0.01	0.26	0.23	0.25	0.01
Exch. K (mg/kg DM)	255	239	245	6.98	185	155	169.5	12.50
Exch. Ca (mg/kg DM)	110	92	101.75	7.68	92	85	88.75	2.98
Exch. Mg (mg/kg DM)	75	62	67.25	5.73	58	42	52.25	7.04
Heavy metal (mg/kg DM)								
Total heavy metal								
Fe	1860	1820	1842.5	17.08	980	890	922.5	40.31
Zn	700	675	688.5	10.28	420	385	401.75	16.89
Mn	402	387	396.75	6.70	205	199	202	2.94
Cu	75	70	72.75	2.21	38	31	34.5	2.88
Co	13.2	12	12.68	0.53	15	10	12.5	2.08
Ni	12	10.8	11.33	0.54				
Cr	51	42	46.5	3.87				
Cd	1.2	0.73	0.92	0.20				
Available heavy metal								
Fe	370	342	357.25	11.58	75	70	72.75	2.21
Zn	162	148	156	6.32	36	28	31.5	3.41
Mn	95	82	89.5	5.80	24	18	21	2.58
Cu	18	14	15.5	1.73	11	9	10	0.81
Co	2.1	1.88	1.98	0.1	2	1.5	1.75	0.20
Ni	3.6	2.9	3.15	0.31				
Cr	17.8	14.5	16.47	1.40				
Cd	0.12	0.08	0.09	0.01				

DM: Dry Matter, Exch: Exchangeable

a one-way analysis of variance (ANOVA) and separated by Duncan's Multiple Range Tests using SAS statistical software[12].

RESULTS AND DISCUSSION

Seed emergence and seedling growth

Seed emergence percentage and period: Effects of sewage sludge combination rate on seed germination percentage and period were not significant. However, both the highest percentage of seed germination (93.3%) and the fast germination period (8.5 days) occurred in the 25% FM+75% SS combination rate (Table 2).

Sewage sludge combination rate positively affected on hypocotyls length, cotyledon length, cotyledon width, shoot length, root length, shoot fresh weight, root fresh weight, shoot dry weight, root dry weight, stem diameter and leaf number per seedling (Table 2).

Hypocotyls length: Sewage sludge applications significantly affected on hypocotyls length. The highest hypocotyls length (22.6 mm) was determined at the 25% FM+75% SS and 50% FM+50% SS combination rates (Table 2).

Cotyledon width and length: The application of 25% FM+75% SS significantly affected the cotyledon width (9.1 mm) and length (26.5 mm) comparing to 100% FM (Table 2).

Shoot length: Sewage sludge applications significantly affected shoot length. The highest shoot length (10.6 cm) was determined at 25% FM+75% SS combination rate when compared to the other treatments (Table 2).

Root length: There were significant differences among sewage sludge applications. Sewage sludge application increased plant root length depending on percentage of the sewage sludge rates. The highest root length (17.3 cm) was obtained from 100% SS combination rate (Table 2).

Shoot and root fresh weight: Forms of sewage sludge application significantly influenced shoot and root fresh weight of the seedlings. The highest shoot weight (5.1 g) was determined at 25% FM+75% SS combination rate while the highest root fresh weight (1.45 g) was designated at 100% SS combination rate (Table 2).

Shoot and root dry weight: Sewage sludge applications significantly affected shoot and root dry weights of the plant seedlings. Shoot (0.53 g) and root (0.095 g) dry weights were higher at 25% FM+75% SS combination than those of the other applications (Table 2).

Stem diameter: Sewage sludge applications significantly were effective on stem diameter. The highest stem diameter (3.1 mm) was determined at the 25% FM+75% SS combination when compared to the other treatments[7].

Leaf number per seedling: Rates of sewage sludge application significantly affected leaf number per seedling. Leaf numbers (14.50) of pepper seedlings were higher at 25% FM+75% SS combination rate than those of the other applications (Table 2).

Plant mineral contents

Nitrogen (N) content: SS and FM combinations had significant effects on N contents of plant. Sole FM (100%) applications had the highest nitrogen (3.55 g 100 plant^{-1}) contents when compared to all of the other treatments. Plant nitrogen content decreased with increasing sewage sludge applications rate (Fig. 1).

Potassium (K) content: The K contents of plants were also significantly affected from the treatments (Fig. 1). The K content (6.62 g 100 g plant^{-1}) of plants

Table 2: Effects of sewage sludge combination rate on seed germination and seedling growth of pepper

	40:20:40 Soil (S), Pumice (PM) and fertiliser mixture [Farmyard Manure (FM)+Sewage Sludge (SS)]				
Properties*	100% FM	75% FM+0.25 SS	50% M+50%SS	25% FM+75% SS	100% SS
Emergence (%)	84.400±4.20ns	80.000±4.12ns	91.100±4.10ns	93.300±3.89ns	91.100±4.00ns
Germination period (day)	9.400±1.14ns	9.400±1.72ns	9.400±0.98ns	8.500±0.86ns	8.700±0.85ns
Hypocotyls length (mm)	16.100±1.1b	20.000±1.4a	22.600±1.2a	22.600±1.4a	21.300±1.3a
Cotyledon length (mm)	17.200±1.3b	22.000±1.1ab	25.700±1.3a	26.500±1.4a	27.100±1.4a
Cotyledon width (mm)	5.600±0.98c	7.800±0.84b	8.500±0.92ab	9.100±0.88ab	9.200±1.00a
Shoot length (cm)	2.700±0.2d	4.300±0.4c	7.900±0.5b	10.600±0.6a	8.800±0.9b
Root length (cm)	2.900±0.2c	5.000±0.5c	9.600±0.3b	12.800±0.8b	17.300±1.1a
Shoot fresh weight (g)	0.200±0.09c	0.600±0.05c	2.500±0.06b	5.100±0.8a	4.200±0.8a
Root fresh weight (g)	0.030±0.01c	0.130±0.04c	0.550±0.03b	1.340±0.04a	1.450±0.05a
Shoot dry weight (g)	0.020±0.009c	0.070±0.002c	0.250±0.005c	0.530±0.004a	0.460±0.004a
Root dry weight (g)	0.006±0.001c	0.015±0.002c	0.048±0.003b	0.095±0.003a	0.091±0.003a
Stem diameter (mm)	1.200±0.05e	1.600±0.03d	2.400±0.06c	3.100±0.04a	2.800±0.03b
Leaf number/seedling	4.830±0.8c	7.170±0.7c	11.330±0.6b	14.500±0.5a	12.830±0.5a

*: Values are the significance tested in rows, significantly important according to p>0.01, ns: not significant

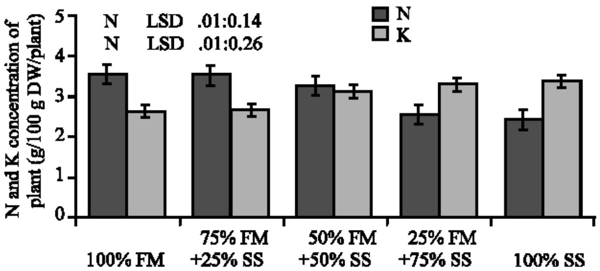

Fig. 1: Effects of sewage sludge in compensation for mineral fertilizer on the N and K content of pepper seedling

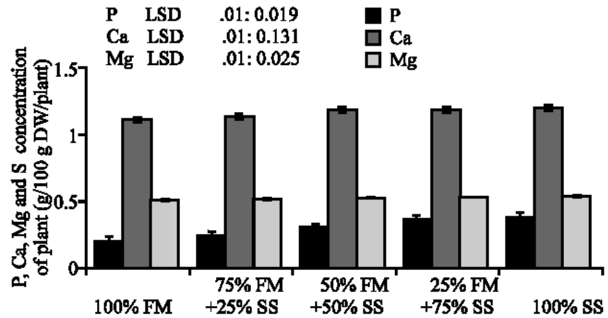

Fig. 2: Effects of sewage sludge in compensation for mineral fertilizer on the P, Ca and Mg content of pepper seedling

in 100% SS treatment was the highest when compared to the other treatments. SS and FM combinations (50%FM+50% SS, 25%FM+75% SS and 100% SS) had similar effects on K contents of plant.

Phosphorous (P) content: SS application significantly affected P contents of plant. The highest phosphorus content was obtained in the 100% SS treatments (0.40 g 100 plant^{-1}) (Fig. 2).

Calcium (Ca) content: Effects of sewage sludge and farmyard Ca contents of plants were not statistically significant (Fig. 2). However the highest Ca content (126 g 100 g plant^{-1}) was obtained in the 100% SS application.

Magnesium (Mg) content: Similarly, SS and FM combinations were not significantly effective on Mg contents of plants (Fig. 2). But, the Mg amounts of treatments in 100%SS treatment were also higher than those of the other treatments.

Manganese (Mn) content: FM and SS combinations significantly affected Mn contents of plants. The Mn content of 100% SS (78.33 mg kg^{-1}) treatment was the highest when compared to all of the other treatments (Fig. 3).

Zinc (Zn) content: The Zn contents of plants were also significantly affected from SS and FM combination (Fig. 3). The Zn contents of 100% SS was the highest when compared to all of the other treatments.

Capper (Cu) content: The Cu contents of plants were significantly influenced from SS and FM combinations (Fig. 3). The Cu contents of treatments of 100% SS (18.05 mg kg^{-1}) and 25% FM+75% SS (13.00 mg kg^{-1}) were higher than those of the other treatments.

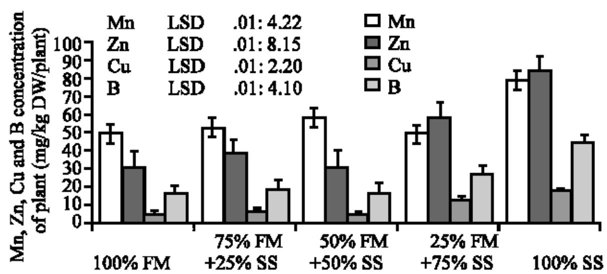

Fig. 3: Effects of sewage sludge in compensation for mineral fertilizer on the Mn, Zn, Cu and B content of pepper seedling

Boron (B) content: The B contents of plants were also significantly affected from SS and FM combination application (Fig. 3). While the highest B content of plants (39.11 mg kg^{-1}) was obtained from the 100% of sewage sludge application, the lowest B content was realized in 100% FM treatments.

Molybdenum (Mo) content: SS and MF applications significantly affected Mo contents of plants. Molybdenum content of plant in the 100%SS treatment had a greater value (1.91 mg kg^{-1}) than the other treatments (Fig. 4).

Cadmium (Cd) content: The Cd contents of plants were significantly affected from SS and FM combination application (Fig. 4). The Cd contents of treatments of 100% SS (0.34 mg kg^{-1}) treatment were higher than those of the other treatments. Plant Cd content increased with increasing sewage sludge applications rate.

FM and SS applications to plant growth media at different ratio affected the seedling growth parameters of peppers. The sewage sludge used in this study was suitable for plant breading purposes in terms of the

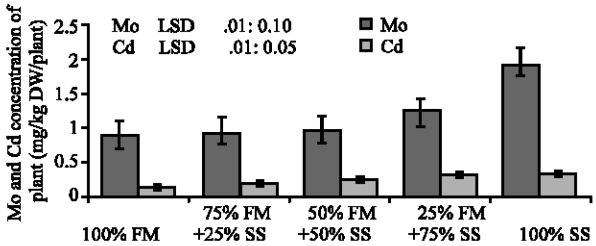

Fig. 4: Effects of sewage sludge in compensation for mineral fertilizer on the Mo and Cd content of pepper seedling

investigated heavy metal contents. Farmyard manure and SS application at 25%FM+75% SS ratio in the growth media mixture were effective combination in seedling parameter, but 50% FM+50% SS ratio was suitable for heavy metal contents of plant. Present results were in good agreement with many researchers[13-16].

The plant development parameters and K, P, Mn, Zn, Cu, B, Mo and Cd contents of pepper were increased with increasing SS applications but this parameter were decreased with increasing FM applications. This can be explained by organic matter reduce the solubility of heavy metal by means of organic-metal chelating properties. There are so many research results supporting present results presented here[3,13,17-19].

Seedling development period is very important for all plant species produced seedling material in terms of quantity and quality on yield. Structure of growth medium and addition of mineral nutrient contents are also important for seedlings. Therefore, SS is important and useful for seedling development due to its organic and mineral contents. In present results, combination of 50% FM+50% SS increased pepper seedling development performance and mineral nutrient contents of plant and did not have a toxic effect on development of seedling performance. So, this ratio will be the most convenient substrate to use, for economical and environmental point of view.

As a result, SS sludge could be a very important alternative of mineral fertilisers in pepper seedling growth. Moreover, the heavy metal toxicity and notion of risk perception has to be clarified and measured. Heavy metal toxicity and accumulation risk in plants are reduced by soil, pumice and organic matter mixtures. In fact, our results show that the farmer should find a long-term advantage in sludge disposal and at the same time satisfy consumer demands for food safety. Meanwhile, in the long run, questions of soil fertility and protection of food chain are raised simultaneously.

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