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Leaf and Seed Micronutrient Accumulation in Soybean Cultivars in Response to Integrated Organic and Chemical Fertilizers Application

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Abstract: Plant nutrients can be influenced by organic materials of soils. An experiment was conducted to evaluate the effect of organic amendments on elements uptake by soybean cultivars in a silty loam soil in Mazandaran province, Iran. The experiment was carried out in split plot based on randomized complete block design with three replications in 2006. Main plots were included 8 fertilizer treatments consisted of 20 and 40 Mg ha^{-1} Municipal Solid Waste Compost (MSW), Vermicompost (VC) and Sewage Sludge (SS) which enriched with 50% chemical fertilizers needed by soil, only chemical fertilizer treatment and control. Sub plots consisted of three genotypes of soybean (032, 033 and JK). Grain yield was determined and soybean leaves and seeds were digested and analyzed for Mn, Cu, Zn and Fe. Results showed that yield and elements content in soybean leaves and seeds (Mn, Cu, Zn and Fe) were influenced by all treatments. The 40 Mg ha^{-1} of sewage sludge enriched with chemical fertilizers produced maximum grain yield. Different soybean cultivars had also significant differences in terms of leaf and seed micronutrients accumulation. Maximum grain yield was observed in JK and 033. Mean comparisons showed that interaction effects of fertilizer and cultivar had significant differences on Mn, Cu and Fe content in soybean leaves, so that the maximum Cu content was observed in 032 cultivars with 40 Mg ha^{-1} enriched sewage sludge and municipal waste compost. Also the highest amount of Fe was obtained for JK cultivar when the 40 Mg ha^{-1} of municipal compost was used. Among different mentioned traits, Fe and Cu content in leaf and seed and Zn content in leaf had a positive and significant correlation with grain yield.

Key words: Soybean, yield, micronutrients, compost, vermicompost, sewage sludge

INTRODUCTION

Soil organic matter (SOM) is universally recognized to be essential for a healthy and productive soil. Among the several agronomic and environmental functions exerted by SOM, nutrient cycling, water retention and drainage, soil susceptibility to contamination and erosion and crop resistance to pests and diseases are those especially dependent on SOM quality and quantity (Rees *et al*., 2001; Magdoff and Weil, 2004). Amount of organic wastes produced by farms, food industries and municipalities including animal manure, municipal Sewage Sludge (SS) and urban solid waste, has also increased (Brebbia *et al*., 2004). Furthermore large volumes of organic waste is generated by the textile industry and released into the environment (Kaushik and Garg, 2003). Concerns about environmental quality have led to the introduction of alternative disposal methods such as the use as nutrient source for plants and as soil conditioners. Municipal Waste Compost (MWC) provides an input of readily available plant nutrients, stimulates microbial activity and contributes to maintaining of nutrient and organic matter pools. The fertilizer value of MWC can be significant but varies considerably depending on origin and processing prior to application (Peterson *et al*., 2003). Sewage sludge composting is being increasingly considered by many municipalities throughout the world because it has several advantages over other disposal strategies. Additionally, the application of composts to agricultural soils has many advantages which include providing a whole array of nutrients to the soil (Gonzalez *et al*., 1992; Sikora and Enkiri, 1999; He *et al*., 2000; Chodak *et al*., 2001; Tejada *et al*., 2001).

The primary plant nutrient associated with sewage sludge is nitrogen (N), however, sludges also contribute significant amounts of other macro and micronutrients (Shober *et al*., 2003; Sims, 1990; Warman, 1986; Zebarth *et al*., 2000). Boron, Fe, Mn, Cu, Zn, Mo and Cl are plant essential micronutrients that are present in sewage sludge and sewage sludge compost, yet some of

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these elements can also be toxic to plants and animals when applied in excessive amounts (Warman and Termeer, 2005). Many researchers have studied the effects of applied sewage sludge on the levels of Cu and/or Zn in corn (Cajuste *et al*., 2000; Cripps *et al*., 1992; Reddy *et al*., 1989). Along with Cu and Zn, other workers have also evaluated Mn (Kiemnec *et al*., 1990; Lutrick *et al*., 1982; Ramachandran and De Souza, 1998) and Mn and Fe (Hernandez *et al*., 1991; Juste and Solda, 1985). Soon *et al*. (1980), Warman (1986) and McBride and Evans (2002) also reported on Mn and Tiffany *et al*. (2000) and Zebarth *et al*. (2000) also evaluated Fe and Mn. Investigations of plant Cu and Zn following sludge compost applications are relatively recent (Sims, 1990; Pichtel and Anderson, 1997; Warman and Termeer, 1996; Wen *et al*., 1999), with fewer studies involving corn (Cajuste *et al*., 2000) or forage compared to other crops. The application of sewage sludge to agricultural land usually increases the Cu and Zn concentrations of amended plants (Warman and Termeer, 2005). The National Research Council (1980) has set the domestic animal mineral tolerance level for Cu at 100 mg Cu kg^{-1} feed (except for sheep, which is set at 25 mg Cu kg^{-1} feed) and for Zn at 300 mg Zn kg^{-1} feed (except for Japanese quail, which is set at 125 mg Zn kg^{-1} feed). Based on a review of the literature, Chang *et al*. (1992) described a methodology for establishing phytotoxicity criteria for Cr, Cu, Ni and Zn from agricultural land application of municipal sewage sludges; they found that the Cu concentration in corn did not rise above the 25 mg Cu kg^{-1} feed quality limit even when 1500 kg Cu ha^{-1} was applied. However, the Zn content in corn leaf tissue can exceed the 300 Mg kg^{-1} limit if high amounts of Zn are applied by sewage sludge (Hinesly *et al*., 1984; Lutrick *et al*., 1982). Chang *et al*. (1992) recommended that up to 3500 kg Zn could be applied per hectare without adverse effects on plant growth; however, there has been strong criticism of these recommendations by Schmidt (1997) and others. Several researchers have shown that the Cu and Zn applied in sewage sludge remained in the plow layer of the soil and does not leach downward (Cripps *et al*., 1992). Kanal and Kuldkepp (1993) compared eight organic treatments including three composts, with and without additional NPK. Their study revealed that some organic amendments were better than others and that additional NPK from chemical fertilizer improved overall potato production. In another experiment, application of sludge amendment (80, 130 and 160 Mg ha^{-1}) increased the average dry weight of sunflower plants (*Helianthus annus* L.) compared to unamended soil (Morera *et al*., 2002). Yield of maize and barley also enhanced as a result of sludge application (Hernandez *et al*., 1991). Use of sewage sludge resulted in more robust plants of *Linium usitatissimum* with faster development and greater biomass production (Tsakou *et al*., 2002). In contrast, Moreno *et al*. (1997) reported negative effects of sludge amendment on yield of *Lactuca sativa*.

The objectives of this study was to evaluate the effect of organic fertilizers enriched with chemical fertilizer on grain yield of soybean and Mn, Cu, Zn and Fe accumulation in soybean seed and leaves.

MATERIALS AND METHODS

General description: The experimental site was located in the North of Iran, in the country of Sari. Soil and organic amendments characteristics at the beginning of the experiment are shown in Table 1 and 2. These soils originated from silty and clay mineral parent material. The soils were sampled on 10 June 2006 between soybean rows, from the 0 to 30 cm layer. According to Table 2, the highest K, Cu and Zn content were observed in MSW compost.

Experimental design and procedures: The experiment was carried out as split plot based on randomized complete block design with two factors and three replications. Main plots were included 8 fertilizer treatments consisted of one rate of chemical fertilizer (potassium sulphate and triple super phosphate (75 kg ha^{-1}) based on soil test recommendations and two rates (20 and 40 Mg ha^{-1}) of MSW compost, SS and VC enriched with half chemical

Table 1: Chemical properties of the soil used in the study (g kg^{-1} dry soil)

Characteristic[a]	N (%)	P	K	Mn	Cu	Zn	Fe	C:N ratio	pH
		(mg kg^{-1})							
Soil	0.23	14.56	278.05	13.96	5.57	1.02	58.47	10.95	7.52

[a]: Data refer to dry matter (105°C) of surface soil (0-30 cm) sieved at <2 mm

Table 2: Chemical properties of the municipal solid waste (MSW) compost, vermicompost (VC), sewage sludge (SS) used in the study

Treatments	N (mg kg^{-1})	P (%)	K	Mn	Cu	Zn	Fe	C:N ratio	pH
			(mg kg^{-1})						
MSW	5832.12	0.45	8485.76	251.96	362.18	766.39	7154.81	11.14	7.41
VC	1833.91	0.62	6228.02	351.60	12.92	51.71	5465.35	11.45	8.05
SS	1993.85	0.43	4893.90	235.64	57.00	471.49	16542.11	7.80	7.44

fertilizer needed by soybean based on soil test recommendation. Sub plots consisted of three genotypes of soybean (032, 033 promising lines and JK cultivar). All of the MSW compost, SS, VC and chemical fertilizer were broadcast by hand and immediately mixed into the soil using a rototiller a day before planting. Plot sizes for crop were 3.0×4.0 m with 5 rows of crops per plot. Space between row and within row was 50 and 6 cm (providing 250 plants per plot), respectively. Sowing in 10 May was done by hand after tilling with a rototiller several times to a depth of about 30 cm. Weeds were controlled by hand weeding using a hoe whenever necessary.

Plant sampling and laboratory analysis: Soybean grain yield was determined by combining a swath in 1×2 m spacing in the center of each split plot. Grain samples were then oven dried (60°C for 48 h) to calculate yield on a dry-matter basis. At flowering initiation, leaf samples were prepared from eight whole plants from each plot. Vegetative samples then were transported in a cool box to the laboratory the same day of sampling washed with distilled water, put in paper bags and oven dried at 65°C for at least 2 days. Once dry, the plant parts were weighed to determine total dry matter. Also grains were hand-harvested in harvest date and dried at 65°C for at least 48 h. Metal contents were determined by Atomic Absorption Spectrometry after calcinations of the sample (0.5 g) at 550°C and solution of the ashes in concentrated HCl (38%) and further dilution to 50 cm^3. Tissue N was determined by digestion in H_2SO_4-H_2O_2. Digests were distilled with 10 M NaOH into boric acid and quantitatively titrated with HCl. The digested samples were analyzed micro element (Mn, Cu, Zn, Fe) and Atomic absorption (Spectra aa-Australia).

Statistical analysis: All data were subjected to Analysis of Variance (ANOVA) using the PROC GLM function of SAS statistical program (SAS, 1997). When there was a significant ($p<0.05$) treatment effect means were compared using Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSION

Grain yield: Results showed that 40 Mg ha^{-1} of sewage sludge enriched with chemical fertilizers (T_6) produced significantly higher ($p<0.05$) grain yield than the other treatments also (averaged 3.8 t ha^{-1}), the 20 Mg ha^{-1} of sewage sludge enriched with chemical fertilizers treatment produced higher crop yield compared to the chemical fertilizers treatment, while no significant differences were detected among both the levels of vermicompost treatment enriched with chemical fertilizers and chemical fertilizers alone (data not shown). Several studies revealed that an increase in soybean yield in three years with compost could be attribute to more favorable plant N status from N mineralization because several studies have presented inconsistent soybean yield responses from in-season N application (Wesley *et al*., 1998; Freeborn *et al*., 2001). While Warman and Havard (1996) reported that in two out of 3 years, conventionally grown (NPK fertilized) potatoes yielded higher than organically grown (compost fertilized) potatoes. With regard to different soybean cultivars, the JK and 033 cultivars produced significantly higher grain yields compared to 032 (Table 3).

Table 3: Mean comparison of study traits in different fertilizer amounts and cultivars

	Seed (mg kg^{-1})				Leaf (mg kg^{-1})			
Treatments	Mn	Cu	Zn	Fe	Mn	Cu	Zn	Fe
Fertilizer (T)								
T_1	19.7±2.1b	19.7±1.1ab	61.7±4.1abc	97.4±18.0a	28.80±2.0bc	14.3±1.7bc	57.50±5.5bcd	206.6±11.2b
T_2	22.1±2.6ab	19.8±0.6ab	60.1±3.7bc	99.8±24.4a	28.10±2.0bc	12.6±1.2cd	54.00±6.0cd	208.2±8.7b
T_3	20.3±1.4ab	19.4±1.3ab	61.3±6.6abc	93.8±16.6a	29.50±2.2bc	15.1±0.4ab	59.40±10.3bc	209.2±9.0b
T_4	20.4±1.7ab	20.9±1.1a	64.8±4.7a	100.6±19.9a	30.90±2.1ab	15.6±1.6a	62.60±4.7ab	265.6±42.8a
T_5	22.3±3.2a	20.3±2.1ab	61.2±5.5abc	101.3±24.2a	29.70±3.4bc	13.4±1.bcd	58.00±6.8bcd	216.4±15.4b
T_6	21.1±1.6ab	20.8±1.5a	63.5±4.7ab	100.6±17.6a	33.17±7.4a	15.6±82.3a	67.60±8.8a	219.9±14.6b
T_7	19.9±1.5b	18.4±1.7b	58.6±4.7c	90.0±16.9ab	27.70±5.6c	12.4±1.8b	51.50±9.2d	200.8±24.4bc
T_8	19.1±1.4b	19.2±2.1ab	60.0±4.3bc	76.9±3.5b	26.70±3.1c	11.5±1.8d	52.07±4.3d	183.8±19.7c
Cultivar (C)								
Jk	19.1±1.7c	19.7±1.4b	59.2±2.3b	84.9±8.1b	26.80±2.0b	13.6±1.5b	56.60±7.8b	221.3±41.0a
032	20.5±2.3b	19.2±2.1b	59.5±3.9b	90.9±20.9b	31.30±5.2a	13.0±3.1ab	55.90±6.9b	209.7±21.1b
033	21.9±4.3a	20.6±0.0a	65.4±3.0a	109.3±19.3a	29.90±3.1a	14.3±1.4a	61.10±10.1a	210.4±23.3b
Significance								
T	ns	ns	ns	*	**	**	**	**
C	**	**	**	**	**	**	*	*
T×C	ns	ns	ns	ns	**	**	ns	**
CV (%)	7.85	6.35	5.63	13.15	8.87	6.10	11.47	7.20

*Means with similar letter(s) are not significantly different at 5% level of probability (DMRT),T_1: 20 Mg ha^{-1} municipal compost+50% chemical fertilizer, T_2: 20 Mg ha^{-1} vermicompost+50% chemical fertilizer, T_3: 20 Mg ha^{-1} sewage sludge+50% chemical fertilizer, T_4: 40 Mg ha^{-1} municipal compost+50% chemical fertilizer, T_5: 40 Mg ha^{-1} vermicompost+50% chemical fertilizer, T_6: 40 Mg ha^{-1} sewage sludge+50% chemical fertilizer, T_7: Chemical fertilizer (Potassium sulphate, Triple superphosphate (75 kg ha^{-1}) and T_8: Control (without chemical or organic fertilizer), ns: not significant

Table 4: Interaction effects means comparison of fertilizer and cultivar on study traits

	Seed (mg kg^{-1})				Leaf (mg kg^{-1})			
Treatments	Mn	Cu	Zn	Fe	Mn	Cu	Zn	Fe
T_1V_1	17.4±0.0f	19.6±0.1a-d	18.0±1.6c-e	93.1±8.8b-g	28.10±1.8b-g	15.2±1.5cd	57.6±8.1c-e	200.8±7.8d-g
T_1V_2	20.1±0.5b-f	18.6±1.0b-e	58.1±1.2d-g	89.8±10.1c-g	29.60±3.1b-f	12.6±1.2f-h	55.9±6.8c-e	215.0±16.5d-f
T_1V_3	21.5±2.4b-e	20.8±1.0ab	66.0±3.8ab	109.4±28.3a-d	28.90±1.2b-g	15.1±1.0cd	59.0±1.9b-e	204.1±0.2d-g
T_2V_1	18.7±1.6d-f	19.2±0.6a-e	59.4±3.4b-g	81.8±1.0e-g	26.30±1.6d-g	11.9±0.0h	50.9±7.5d-e	203.7±5.6d-g
T_2V_2	21.2±2.8b-e	20.1±0.5a-d	57.0±2.5e-g	100.0±40.9a-f	29.20±2.1b-g	12.2±1.2gh	54.5±1.2c-e	204.8±0.0d-g
T_2V_3	23.2±1.2ab	20.1±0.5a-d	63.6±2.2a-e	117.8±18.8a	28.70±1.6b-g	13.7±1.2d-g	56.5±7.8c-e	216.1±11.5b-e
T_3V_1	19.6±2.1c-f	19.0±1.6a-e	56.0±2.2fg	81.4±2.5e-g	27.20±1.3c-g	14.8±0.6c-e	57.4±7.1c-e	221.0±0.0d-g
T_3V_2	20.9±0.6b-e	18.9±0.6a-e	62.5±6.1a-f	87.2±9.8d-g	30.20±3.1b-f	15.3±0.4cd	49.8±1.0e	203.9±3.4d-g
T_3V_3	21.5±1.5b-e	20.3±1.6a-c	65.4±7.8a-c	112.7±13.3a-c	31.10±0.8b-d	15.1±0.0cd	71.1±4.9ab	202.6±0.0d-g
T_4V_1	19.5±1.7c-f	20.0±0.6a-d	61.0±1.8b-g	88.0±14.0d-g	30.34±0.0b-f	13.7±0.6d-g	63.5±4.3b-d	320.7±0.0a
T_4V_2	20.6±0.6b-e	21.4±4.1a	64.3±0.6a-d	98.0±15.4a-g	30.00±1.6b-f	17.3±0.0ab	62.1±0.7b-e	240.9±21.9b
T_4V_3	21.0±2.7b-e	21.4±1.2a	68.6±6.6a	116.0±23.3ab	32.50±3.2b	15.9±0.6bc	62.1±8.2b-e	235.1±0.0bc
T_5V_1	19.1±1.4c-f	20.7±1.8ab	61.2±0.3b-g	84.0±8.9eg	27.00±0.1c-g	13.3±2.5e-h	58.8±11.4b-e	212.5±10.9d-f
T_5V_2	22.0±1.9bc	19.2±0.6a-e	57.4±0.6e-g	103.6±34.3a-d	30.70±2.5b-e	12.2±1.2g-h	56.3±3.9c-e	216.7±25.8b-d
T_5V_3	25.7±2.0a	21.0±3.5ab	65.0±2.5a-c	116.3±17.5ab	31.40±4.9b-d	14.6±1.6c-e	58.8±5.9b-e	219.9±11.2b-d
T_6V_1	20.1±1.0b-f	20.7±1.8ab	59.2±0.6b-g	89.8±9.1c-g	25.40±.9f-g	14.4±0.6c-e	60.5±3.4b-e	225.3±3.7b-d
T_6V_2	21.2±1.6b-e	20.7±2.1ab	63.2±2.1a-e	97.0±23.0a-g	42.20±0.0a	18.4±0.0a	65.5±3.2a-c	208.4±2.7c-g
T_6V_3	21.8±2.1b-d	21.0±1.2ab	68.0±5.1a	114.9±11.1ab	31.70±1.5bc	14.0±1.8d-f	76.9±8.6a	225.9±23.9b-d
T_7V_1	19.2±1.4c-f	18.0±1.6c-e	57.0±0.6e-g	82.5±7.2e-g	25.60±1.6e-g	13.7±0.6d-g	54.1±11.2c-e	200.4±14.5d-g
T_7V_2	19.2±1.1c-f	17.8±1.6de	55.4±0.3g	77.4±2.1f-g	19.00±8.2b-g	10.1±0.3i	50.5±10.9d-e	203.8±2.9d-g
T_7V_3	21.3±1.0b-e	19.6±1.8a-d	63.2±6.0a-e	110.1±12.5a-d	28.50±4.9b-g	13.3±0.8e-h	50.0±9.0e	198.3±41.5d-g
T_8V_1	18.8±1.2c-f	20.7±1.5ab	58.9±0.1c-g	79.1±4.9e-g	24.20±0.6g	12.2±1.2gh	49.8±4.3e	185.9±18.3e-g
T_8V_2	18.5±2.1ef	16.9±1.6e	57.8±1.8d-g	74.1±1.0g	29.60±3.8b-f	9.9±2.3i	52.5±5.9c-e	184.2±23.8fg
T_8V_3	20.1±0.5b-f	20.1±0.5a-d	63.2±6.8a-e	77.5±2.6fg	26.30±1.2d-g	12.4±0.9f-h	53.8±3.1c-e	181.3±25.1g

Means with similar letter(s) are not significantly different at 5% level of probability (DMRT), T: Different fertilizer treatments V_1: JK, V_2: 032 and V_3: 033

Seed Mn: All of organic fertilizers enriched with chemical fertilizer except 20 Mg ha^{-1} municipal solid waste treatment increased the Mn content of the soybean seeds. Among genotypes Mn concentrations in seeds were highest in the 033 cultivar (Table 3). The application of AES to the corn plots resulted in higher tissue Mn compared to synthetic fertilizer or compost applications. Since Mn availability is strongly affected by the soil oxidation-reduction potential, the response may be the combined effect of high Mn content in the Aerobically-digested Sludge (AES) and low oxygen conditions in the sewage sludge clods which lowered the soil redox potential, causing an increase in stover Mn (Warman and Termeer, 2005).

Leaf Mn: Application of T_4 (40 Mg ha^{-1} MWS+50% fertilizer) and T_6 (40 Mg ha^{-1} SS+50% fertilizer) to the soybean plots increased the leaf Mn concentration compared to chemical fertilizer and other organic fertilizers plots. Mn concentrations in soybean leaves were highest in 033 and 032 cultivars (Table 3). The Mn content of the soybean leaves was highest in 032 cultivar when the 40 Mg ha^{-1} sewage sludge enriched with 50% chemical fertilizer were applied (Table 4). Warman and Havard (1998) revealed that Phosphorus, Mg and Na were higher in the tubers of organically grown potatoes and B and Fe were higher in the leaves; tuber Mn and leaf N and Mg were higher in conventionally grown potato leaves. As indicated above, tuber Mn and leaf Fe were the only micronutrients statistically influenced by treatments; however, leaf Mn and Cu were always higher in the conventionally grown potatoes although the treatments had no effect on soil Mn and soil Cu was higher in the compost-amended soil. Mn concentrations in shoots and roots of plants, however, were lower when grown in sewage sludge amended soil as compared to those grown in un amended ones (Singh and Agrawal, 2007).

Seed Cu: Results in Table 3 showed that in all of organic fertilizers treatments enriched with chemical fertilizer and control the Cu content of the soybean seeds was highest than chemical fertilizer. Cu concentrations in soybean seeds was highest in 033 cultivar compared to other genotypes (Table 3). The Anaerobic septic sludge (ANS) used had a copper content of 848 mg Cu kg^{-1}, which was the highest copper content, all the amendments Septic sludge applications provided 13.7 kg Cu ha^{-1} for the two years, well below the NSDEL guideline of 150 kg ha^{-1}; however, this application still increased the grass forage Cu concentration. The sludge compost applied had copper content of 238 mg Cu kg^{-1}, but the applications did not increase the Cu concentrations in either the grass forage or corn tissue. The Cu content of the AES amended corn stover was significantly greater than the compost and fertilizer amended corn in 1994, but was not significantly different possibly due to high replicate variability (Warman and Termeer, 2005).

Leaf Cu: The highest leaf Cu content were observed in the 20, 40 Mg ha^{-1} SS and the 40 Mg ha^{-1} MWS in combination with 50% chemical fertilizer treatments (Table 3). Soon *et al.* (1980) reported small increases in the Cu content of bromegrass and corn grain following five years of sewage sludge applications of as high as 31 kg Cu ha^{-1}, while Kiemnec *et al.* (1990) recorded only modest increases in sweet corn leaf and grain Cu content after seven years of sewage sludge applications of as high as 62 kg Cu ha^{-1} to a silt loam soil. Cu concentrations in soybean leaves were highest in 033 line (Table 3). Also, the Cu content of the soybean leaves was highest in 032 cultivar when the 40 Mg ha^{-1} municipal solid waste compost and sewage sludge enriched with 50% chemical fertilizer were applied (Table 4).

Seed Zn: All of organic fertilizers enriched with chemical fertilizer except 20 Mg ha^{-1} VC applications produced higher seed Zn content compared to the chemical fertilizer and Control at three the soybean cultivars. Among cultivars, the seed Zn concentrations were highest in 033 line (Table 3). Applications of both ANS and compost increased zinc concentrations in the grass forage tissue, but the increase was greater from the ANS, which is related to its total Zn content (Warman and Termeer, 2005).

Leaf Zn: The Zn content of the soybean leaves was highest in the 40 Mg ha^{-1} municipal solid waste compost and sewage sludge enriched with chemical fertilizer plots (averaged 62.6 and 67.6 mg kg^{-1} dry weight, respectively). Although the long term studies of Soon *et al.* (1980) and Kiemnec *et al.* (1990) indicated relatively small increases in corn Zn content, very high Zn applications in sewage sludge can increase tissue Zn above 300 mg Zn kg^{-1} feed (Warman and Termeer, 2005). Zn concentrations in soybean leaves were highest in 033 line (Table 3). Heavy metal (Zn, Ni, Cd and Cu) accumulation in plant tissues was reported in plants grown on sludge-amended soil (Moreno *et al.*, 1997). Zn, Pb, Cd, Ni, Cr and Cu concentrations in shoots and roots of plants grown in sewage sludge-amended soils were significantly higher as compared to those in unamended soil. Ni, Cd, Pb and Cr concentrations in shoot were higher at 40% than 20% Sewage Sludge Amendment (SSA), whereas Cu and Zn showed a reverse trend of higher concentration at 20% SSA. Cd, Ni, Zn, Cr and Cu concentrations in roots were significantly higher in plants grown at 40% as compared to 20% SSA, however, Pb concentration was higher at 20% then 40% SSA (Singh and Agrawal, 2007). Normally the Zn content increases more substantially than the Cu content. The factors that affect the bioavailability of an element include soil pH, plant species and their cultivars, growth stage, biosolid source, soil conditions and the chemistry of the element (Warman and Termeer, 2005).

Seed Fe: The Fe content of the soybean seeds was higher in organic treatment plots compared to control. The highest Fe content in seed of soybean cultivars belonged to 033 line (Table 3). Although AES and the compost made from it have a high concentration of iron and considerable amounts of Fe were applied by the organic amendments to the forage and corn plots, only the ANS had much of an effect on tissue Fe concentrations (Warman and Termeer, 2005).

Leaf Fe: The 40 Mg ha^{-1} MSW enriched with chemical fertilizer caused the highest concentrations of Fe in the plant leaves compared to other treatments. The highest Fe content in seed of soybean cultivars was observed in JK cultivar (Table 3). The result of ANOVA showed that interaction effects of fertilizer and cultivar had significant differences on Fe content in soybean leaves so that the highest amount of iron was obtained when the 40 Mg ha^{-1} municipal compost enriched with chemical fertilizer for JK cultivar was used. Higher amounts of Fe, Cu and Zn were absorbed in maize and barley grown on sludge-amended soil than those grown on the unamended ones (Hernandez *et al.*, 1991). There was a significant correlation between grain yield soybean and Cu (r = 0.25*), seed Fe (r = 0.28*), Cu (r = 0.36**), Zn (r = 0.44**) and leaf Fe (r = 0.27*) (Table 5).

Table 5: Correlation coefficients of grain yield with studied traits (n = 72)

No.	SOV	1	2	3	4	5	6	7	8	9
1	Seed yield									
2	Seed Mn		0.08							
3	Leaf Mn		0.14	0.20						
4	Seed Cu		0.25*	0.26*	0.10					
5	Leaf Cu		0.36**	0.17	0.39**	0.39**				
6	Seed Zn		0.14	0.30**	0.23**	0.51**	0.40**			
7	Leaf Zn		0.44**	0.15	0.39**	0.37**	0.42**	0.30**		
8	Seed Fe		0.28*	0.49**	0.18	0.50**	0.32**	0.57**	0.26*	
9	Leaf Fe		0.27*	0.10	0.21	0.13	0.22	0.08	0.31**	0.07

* and **Significant at 5 and 1%, respectively

CONCLUSIONS

In conclusion, we observed that a mixture of 40 Mg ha^{-1} sewage sludge and inorganic fertilizers produce higher yields than other fertilizer treatments. The 40 Mg ha^{-1} of sewage sludge enriched with half chemical fertilizer increased the grain yield and all of seed micronutrients concentration compared to other fertilizer treatments. Also the 40 of municipal waste compost enriched with half chemical fertilizer increased the all of leaf micronutrients concentration compared to other fertilizers. Therefore, based on the results of the current experiment, organic fertilizers which enriched with chemical fertilizer can be used in place of inorganic fertilizer alone to supply Mn, Cu, Fe and Zn of soybean grown. This can help recycle plant nutrients and thus reduce environmental degradation associated with the disposal of MSW, SS and VC to landfills. The result of mean comparisons showed that interaction effects of fertilizer and cultivar had significant differences on Mn, Cu and Fe content in soybean leaves so that most Cu amounts were observed in 032 line and in 40 Mg ha^{-1} enriched SS and MSW compost enriched with half chemical fertilizer. Also the highest leaf Fe was obtained when the 40 Mg ha^{-1} MSW compost enriched with half chemical fertilizer for JK cultivar was used. The highest leaf Mn was obtained when the 40 Mg ha^{-1} SS enriched with half chemical fertilizer for 032 line was used. Fe and Cu content in leaf and seed and Zn content in leaf had a positive and significant correlation with grain yield.

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