Research in Agriculture
ISSN 2740-4431 (Print) ISSN 2740-444X (Online)
Vol. 1, No. 1, 2016
www.scholink.org/ojs/index.php/ra

Agronomical Performance of High Yielding Cultivar of Eggplant (*Solanum melongena* L.) Grown in Sewage Sludge Amended Soil

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Abstract

A field study was conducted to assess the agronomical performance of eggplant (Solanum melongena cv. Pusa Purple Long Hybrid-F_1) grown in sewage sludge amended soil in rainy and summer seasons during two consecutive years 2012 and 2013. The results showed that sewage sludge significantly ($P < 0.05$/$P < 0.01$) increased the Organic Carbon (OC), sodium (Na^{+}), potassium (K^{+}), calcium (Ca^{2+}), magnesium (Mg^{2+}), Total Kjeldahl Nitrogen (TKN), phosphate (PO_4^{3-}), sulphate (SO_4^{2-}), cadmium (Cd), chromium (Cr), copper (Cu), manganese (Mn) and zinc (Zn) in the soil. The contents of metals were found below the harmful levels prescribed for soils in India. The maximum agronomic performance in the form of plant height, root length, dry weight, chlorophyll content, Leaf Area Index (LAI), yield/plant and biochemical components like total carbohydrate, crude protein and dietary fiber of S. melongena were observed at 50% concentration of sewage sludge in both seasons. The contents of Cd, Cr, Cu, Mn and Zn in S. melongena were increased from 5% to 100% treatments of sewage sludge in both seasons. The order of Contamination factor (Cf) of different heavy metals was Mn > Cr > Cd > Zn > Cu for soil and Cr > Cd > Cu > Mn > Zn for S. melongena plants after treatment of sewage sludge. Thus, use of sewage sludge increased concentration of heavy metals in soil and S. melongena. Hence, sewage sludge can be governed as organic fertilizer in lower proportion to avoid the contamination of soil and S. melongena.

Keywords

agronomical characteristics, Contamination factor, heavy metals, sewage sludge, solanum melongena

1. Introduction

Sewage sludge refers to the residual, semi-solid material that is produced as a by-product during sewage treatment of industrial or municipal wastewater (Zhang et al., 2007; Esteller et al., 2009). Sewage sludge is produced from the treatment of wastewater in sewage treatment plants (Esteller et al., 2009; Kumar & Chopra, 2012; Kumar & Chopra, 2014a). Sewage sludge contains useful concentrations of nitrogen (N), phosphorus (P) and organic carbon (OC) (Mehmet, 2013; Kumar & Chopra, 2014c, 2014e). The availability of N is more dependent on sludge treatment, untreated liquid sludge and dewatered treated sludge releasing nitrogen slowly with the benefits to crops being realized over a relatively long period (Zhou et al., 2012; Kumar & Chopra, 2014d). The organic matter in sludge can improve the water retaining capacity and structure of some soils, especially when applied in the form of dewatered sludge cake (Keeling et al., 1995; Wei, 2002; Mehmet, 2013).

Apart from those components of concern, sewage sludge also contains pathogenic bacteria, viruses and protozoa along with other parasitic helminths which can give rise to potential hazards to the health of humans, animals and plants (Wei & Liu, 2005; Akdeniz et al., 2006). The numbers of pathogenic and parasitic organisms in sludge can be significantly reduced before application to the land by appropriate sludge treatment and the potential health risk is further reduced by the effects of climate, soil-microorganisms and time after the sludge is applied to the soil (Benitez et al., 2001; Zhang et al., 2007).

Sewage sludge application to agricultural soil has been a widely accepted practice during recent years (Naggar & El-Ghamry, 2001; Kumar & Chopra, 2012). Its use in agricultural soil is promoted because it is considered that it will solve not only the problem of disposal but also will increase productivity in agriculture (Pedreno et al., 1996; Kumar & Chopra, 2014a, 2014b). However, negative effects of sewage sludge such as elevated heavy metals like Cd, Cr, Cu, Mn, Pb and Zn levels resulting from the usage of sewage sludge must also be taken into consideration (Barman et al., 2000; Alcantara et al., 2009).

Sewage sludge contains appreciable amounts of N, P, calcium (Ca), magnesium (Mg), potassium (K) and organic matter (Kim et al., 2003). Thus, the agricultural use of sewage sludge may be an alternative option to conventional inorganic fertilizers (Garrido et al., 2005; Casado-Vela et al., 2007; Samaras et al., 2008; Kumar & Chopra, 2013c). Although, sewage sludge provides nutrients for plant growth, its continual use over extended periods can result in the accumulation of heavy metals to levels detrimental to the environment. Sewage sludge also contains heavy metals and organic pollutants which may have adverse effects on crop plants (Dursan et al., 2005; Muchuweti et al., 2006; Fukuoka et al., 2010; Kumar & Chopra, 2014c).

The sewage sludge may contain trace elements at levels injurious to plants and the food chain. Zinc (Zn) and copper (Cu) are essential for plant growth but are, together with Nickel (Ni), toxic to plants above certain thresholds. Lead (Pb) can be injurious to plants in low P acid soils, but Pb added with sewage sludge is usually non-toxic to plants because of the large amount of P also contained in the sludge which can control Pb solubility (Mohammad & Athamneh, 2004; Kumar & Chopra, 2013c). Moreover, the disposal of sewage sludge is a major problem due to increase in urbanization and industrialization, especially in developing countries, and it requires municipal authorities to handle larger volumes of sewage sludge often with limited resources. High energy requirements for sewage sludge incineration, scarcity of landfill sites for the disposal of sewage sludge (Mamo et al., 1999; Perez-Espinosa et al., 1999; Petersen et al., 2003; Sharma et al., 2007). Sludge type and sludge-soil interactions influence the chemical forms of a metal which determine its availability for plant uptake. However, additional plant and soil factors further modify the uptake and the concentration of elements in crops (Singh & Agrawal, 2007; Sridhara et al., 2008; Tamrabet et al., 2009; Kumar & Chopra, 2013a, 2013b).

The recycling of sewage sludge to agricultural land is favoured over disposal through landfill or incineration due to potentially beneficial effects on soil fertility from organic matter and plant nutrients such as N and P in the sludge. Thus, sludge addition usually produced a stimulation of plant growth (Yadav et al., 2002; Sharma et al., 2007; Zhou et al., 2012). Moreover, some sludge components are required for microbial growth and increase the activity of soil microorganisms to enhance the soil fertility (Sridhara et al., 2008; Saxena & Diwakar, 2012). Heavy metals contamination in soils has attracted serious attention in recent years, which poses a considerable hazard to health (Singh & Agrawal, 2007; Sridhara et al., 2008; Togay et al., 2008). Heavy metals such as Zn and Cu are essential nutrients for plants and are present in sewage sludge and sludge compost. Research has shown the effects of the application of sewage sludge on Cu and Zn levels in maize. Sewage sludge promoted an increase in total Zn concentration without becoming excessive for human consumption (Togay et al., 2008; Kumar & Chopra, 2014d).

Eggplant (*Solanum melongena* L.) is identified as one of the most valuable vegetable packed with essential nutrients (Fukuoka et al., 2010; Dar et al., 2014). The eggplant is a delicate, tropical perennial often cultivated as a tender or half-hardy annual in temperate climates (Nunome et al., 2001; Hirakawa et al., 2014). It is being widely cultivated throughout the world in tropical and subtropical climates. Nutritionally, eggplant is low in fat, protein, and carbohydrates. It is rich in dietary fiber, sugar, sodium and potassium (Fukuoka et al., 2010). It also contains important vitamins like A, B_6, C, D and calcium, iron and magnesium. Eggplant is used in the cuisine of many countries (Zhou et al., 2012). It is widely used in its native Indian cuisine, like vegetable, chutney, curry and pickle. Eggplant, due to its texture and bulk, can be used as a meat substitute in vegan and vegetarian cuisine. The juice of eggplant significantly reduces weight, plasma cholesterol levels, and aortic cholesterol content (Elanchezhyan et al., 2008; Prabhu et al., 2009; Chopra et al., 2013; Hirakawa et al., 2014). The disposal of sewage sludge is an environmental problem that cities face today, and the use of these wastes as fertilizers is an issue of debate (Muchuweti et al., 2006; Kidd et al., 2007; Tamrabet et al., 2009; Kumar & Chopra, 2013c). Keeping above in view, the present study was carried out to assess the accumulation of heavy metals in eggplant (*Solanum melongena* L.) grown in sewage sludge amended soil.

2. Materials and Methods

2.1 Experimental Design

The field trials were conducted in the Experimental Garden of the Department of Zoology and Environmental Sciences, Gurukula Kangri University Haridwar, India (29^{o}55'10.81" N and 78^{o}07'08.12" E), to study the agronomical performance of *S. melongena* grown in sewage sludge amended soil. The crop was cultivated in the rainy and summer seasons during the year 2012 and 2013. Seven plots (each plot had an area of 9 × 9 m^2) were selected for seven treatments of sewage sludge *viz.*, 0% (Garden soil as control), 5% (5% sludge + 95% garden soil), 10% (10% sludge + 90% garden soil), 25% (25% sludge + 75% garden soil), 50% (50% sludge + 50% garden soil), 75% (75% sludge + 25% garden soil) and 100% (100% sludge) for the cultivation of *S. melongena*. All the seven treatments were placed within each of the seven plots in a randomized complete block design.

2.2 Collection of Sewage Sludge and Analysis

The sewage sludge samples were collected from municipal sewage treatment plant, Kankhal, Haridwar, Uttarakhand (29°54'2"N 78°8'18"E). The sewage sludge was collected using plastic bags from sludge drying beds located in the treatment plant at the campus. It was brought to the laboratory and analyzed for pH, electrical conductivity (EC), chlorides (Cl^-), Na^+, K^+, Ca^{2+}, Mg^{2+}, total Kjeldahl nitrogen (TKN), phosphate (PO_4^{3-}), sulphate (SO_4^{2-}), Cd, Cr, Cu, Fe, Mn, and Zn following standard methods (Chaturvedi & Sankar, 2006) and used as soil amendment.

2.3 Preparation of Nursery of S. Melongena

Seeds of a high yield variety of *S. melongena*, cv. Pusa Purple Long Hybrid-F_1, were procured from Indian Council of Agriculture Research (ICAR), Pusa, New Delhi, and sterilized with 0.01% Thiram. The nursery was prepared before one month of transplanting of *S. melongena*. For the nursery preparation, a 1 × 1m size of raised nursery bed was prepared with the help of bricks. 5.0 kg of farm yard manure was mixed in the soil and treated with 5% dust of Aldrex @ 75Kg/h. 0.5 g seeds of eggplant were sown in the rows 5 cm apart on 2.5 cm nursery beds (farm yard manure mixed soil) and covered with a thin layer of the soil. The nursery bed was covered with straw mulch till the seeds germinate. The plants were watered as per requirement and other agronomical practices like weeding and hoeing were performed manually till the plants were transplanted in the field.

2.4 Transplantation and Cultivation Practices of S. Melongena

Four weeks old plants of *S. melongena* were planted in the forth week of July 2012 and 2013 for the rainy season crop and in the forth week of April 2012 and 2013 for the summer season crop. Plants of *S. melongena* were transplanted in 6 rows with a distance of 60 × 60 cm between plants (Saxena & Diwakar, 2012). The plants in each plot were irrigated twice in a month with 400 liter of bore well water and necessary agronomical practices were performed. The soil was analyzed prior to planting and after harvest for various physico-chemical parameters like soil texture, bulk density (BD), water holding capacity (WHC), EC, pH, OC, Na^+, K^+, Ca^{2+}, Fe^{2+}, Mg^{2+}, PO_4^{3-}, SO_4^{2-}, TKN, Cd, Cr, Cu, Mn and Zn determined following standard methods (Chaturvedi & Sankar, 2006).

2.5 Study of Crop Parameters

The agronomic parameters of *S. melongena* at different stages (0-110 days) were determined following standard methods for seed germination, plant height, root length, number of leaves, leaves length and crop yield (Saxena & Diwakar, 2012); dry weight (Denison & Russotti, 1997); chlorophyll content (Porra, 2002) and leaf area index (LAI) (Milner & Hughes, 1968). Total carbohydrate, crude protein and dietary fiber in *S. melongena* plants were determined following standard methods (Chaturvedi & Sankar, 2006).

2.6 Extraction of Metals and Their Analysis

For metal analysis 1.0g of air dried sewage sludge, soil or plants were taken in digestion tubes separately. For each sample 3 ml of concentrate HNO_3 was added and digested in an electrically heated block for 1 hour at 145^{o}C. To this mixture 4 ml of $HClO_4$ was added and heated to 240^{o}C for 1 hour. The mixture was cooled and filtered through Whatman # 42 filter paper. The volume was made to 50 ml by adding double distilled water and used for analysis. Metals were analyzed using an atomic absorption spectrophotometer (PerkinElmer, Analyst 800 AAS, GenTech Scientific Inc., Arcade, NY) following methods (Chaturvedi & Sankar, 2006). In this field study, the contamination factor (Cf) was used to determine the contamination of metals in soil and *S. melongena* amended with sewage sludge. The Cf was calculated following formula (Chaturvedi & Sankar, 2006; Kumar & Chopra, 2013c).

2.7 Data Analysis

Data were analyzed with SPSS (ver. 14.0, SPSS Inc., Chicago, Ill.). Data were subjected to one-way analysis of variance (ANOVA). Mean standard deviation and coefficient of correlation (r-value) of soil and crop parameters with sludge concentrations were calculated with MS Excel (ver. 2007, Microsoft Redmond Campus, Redmond, WA) and graphs produced with Sigma plot (ver. 12.3, Systat Software, Inc., Chicago, IL).

3. Results and Discussion

3.1 Characteristics of Sewage Sludge

The physico-chemical and heavy metals characteristics of sewage sludge are shown in Table 1. The sewage sludge parameters were significantly ($P < 0.05/P < 0.01/P < 0.001$) different over concentrations. The sewage sludge was noted alkaline (8.40) in nature (Table 1). During the present study the alkaline nature of the sewage sludge may be likely due to the use of alkalis in the household process. The higher contents of OC (9.20 mg kg^{-1}), Na^+ (518.30 mg kg^{-1}), K^+ (978.50 mg kg^{-1}), Ca^{2+} (825.68 mg kg^{-1}), Mg^{2+} (268.60 mg kg^{-1}), PO_4^{3-} (272.55 mg kg^{-1}), SO_4^{2-} (948.50 mg kg^{-1}), TKN (332.89 mg kg^{-1}), Fe (24.88 mg kg^{-1}), Cd (10.76 mg kg^{-1}), Cr (6.12 mg kg^{-1}), Cu (21.20 mg kg^{-1}), Mn (14.50 mg kg^{-1}) and Zn (30.95 mg kg^{-1}) were noted in 100% sewage sludge. The findings are in line of Khan et al. (2007) who, reported higher contents of OC (12.50 mg kg^{-1}), P (17.40 mg kg^{-1}), TKN (65.00 mg kg^{-1}) and metals Cd (2.30 mg kg^{-1}), Cu (51 mg kg^{-1}), Pb (21.90 mg kg^{-1}) and Zn (441 mg kg^{-1}) in sewage sludge. Moreover, the findings are in accordance with Kumar and Chopra (2013b) who reported slightly lower contents of PO_4^{3-} (215.83), NO_3^{-2} (356.93), Cd (10.24), Cu (18.96), Fe (19.81), Pb (9.33) and Zn (11.25) in sewage sludge.

Table 1. Physico-Chemical Characteristics of Sewage Sludge

Parameter	Sewage sludge concentration (%)						
	0 (GS)[a]	5	10	25	50	75	100
EC (dS m^{-1})	0.24 ±0.03	0.28* ±0.05	0.56** ±0.07	1.14** ±0.04	2.30** ±0.05	3.40** ±0.06	4.63** ±0.07
pH	7.48 ±1.02	7.55ns ±1.00	7.62ns ±1.05	7.74ns ±1.03	8.15ns ±1.04	8.25ns ±1.02	8.40ns ±1.12
OC (mg kg^{-1})	0.44 ±0.02	0.75** ±0.04	1.56** ±0.08	2.28** ±0.12	4.60** ±0.75	6.94*** ±0.66	9.20*** ±0.1.03
Na^+ (mg kg^{-1})	24.50 ±1.55	45.60* ±1.67	85.97* ±1.34	157.34** ±1.44	288.70** ±1.10	396.53*** ±1.12	518.30*** ±1.33
K^+ (mg kg^{-1})	60.20 ±2.10	80.60* ±2.24	160.34* ±2.15	242.60** ±2.34	485.88** ±2.12	732.60*** ±2.09	978.50*** ±2.75
Ca^{2+} (mg kg^{-1})	30.44 ±2.16	70.50* ±2.77	135.90* ±2.44	274.20** ±2.13	454.86** ±2.54	594.70*** ±2.30	825.68*** ±2.10
Mg^{2+} (mg kg^{-1})	3.40 ±1.00	8.60* ±1.07	18.50* ±1.05	56.34** ±1.23	127.40** ±1.78	205.75*** ±2.45	268.60*** ±2.34
TKN (mg kg^{-1})	28.90 ±1.04	32.40* ±2.09	66.80* ±2.11	117.40** ±2.78	162.80** ±2.55	276.30*** ±2.45	332.89*** ±2.10
PO_4^{3-} (mg kg^{-1})	2.65 ±0.14	14.60* ±1.01	25.89* ±1.02	76.87** ±1.96	134.50** ±1.47	202.80*** ±1.23	272.55*** ±1.20
SO_4^{2-} (mg kg^{-1})	77.89 ±0.50	90.30* ±1.21	187.97* ±1.77	352.85** ±2.09	467.40** ±2.10	583.60*** ±3.20	948.50*** ±3.21
Fe (mg kg^{-1})	1.50 ±0.23	1.36* ±0.64	2.80* ±0.96	5.20** ±0.78	12.67** ±0.64	18.09*** ±0.66	24.88*** ±0.70
Cd (mg kg^{-1})	0.53 ±0.06	0.79* ±0.09	1.59** ±0.04	3.38** ±0.08	5.34** ±0.05	8.12*** ±0.05	10.76*** ±0.07
Cr (mg kg^{-1})	0.36 ±0.01	0.40* ±0.02	0.83* ±0.04	1.69** ±0.03	3.04** ±0.02	4.84*** ±0.06	6.12*** ±0.04
Cu (mg kg^{-1})	2.37 ±0.02	2.45* ±0.04	4.93* ±0.09	6.82** ±0.04	10.24** ±0.08	16.95*** ±0.05	21.20*** ±0.08
Mn (mg kg^{-1})	0.54 ±0.01	0.82* ±0.04	1.65** ±0.07	3.75** ±0.06	7.02** ±0.09	11.60*** ±1.00	14.50*** ±1.10
Zn (mg kg^{-1})	2.85 ±0.06	2.90* ±0.09	5.89** ±0.08	12.86** ±0.12	15.96** ±0.19	25.94*** ±0.18	30.95*** ±0.17

Note: Mean ± SE of three values; ns, *, **, *** non-significant or significant at $P \leq 0.05$ or $P \leq 0.01$ or

P ≤ 0.001, ANOVA;

[a] GS = Garden soil (control).

3.2 Effect of Sewage Sludge on Characteristics of Soil

The present study revealed that use of 100% sewage sludge insignificantly (P > 0.05) reduced the water holding capacity (40.00%) and bulk density (1.30 gm cm^{-3}) from their initial values water holding capacity (41.80%) and bulk density (1.31 gm cm^{-3}), respectively. Moreover, water holding capacity and bulk density were negatively correlated (r = -66 and -68, respectively) with all concentrations of sewage sludge in both the cultivated seasons (Table 2). This slight decrease in water holding capacity and bulk density are likely due to the presence of more organic matter in sewage sludge. The findings are in agreement with Kumar and Chopra (2013b) who, reported decrease in water holding capacity and bulk density of the sewage sludge treated soil. At harvest (110 days after sowing of *S. melongena*) there was insignificant change in the soil texture (sandy loam; 60% sand: 20% silt: 20% clay). In the present study, the pH of the soil was turned alkaline to more alkaline (8.67 and 8.74) after amendment with 100% sewage sludge in both seasons (Table 2). Soil pH was significantly (P < 0.05) affected by 50% to 100% treatments of sewage sludge (Table 2).

Table 2a. Effects of Sewage Sludge (SS) Concentration and Season Interaction on Physico-Chemical Characteristics of Soil after Cultivation of *S. Melongena* in both Seasons

Season	× %SS	EC (dS m^{-1})	pH	OC (mg kg^{-1})	Na^{+} (mg kg^{-1})	K^{+} (mg kg^{-1})	Ca^{2+} (mg kg^{-1})	Mg^{2+} (mg kg^{-1})
Rainy	0 (Control)	0.45	7.60	0.52	18.76	164.00	26.70	2.56
		±0.03	±0.10	±0.02	±1.11	±2.10	±1.02	±0.05
	5	1.28ns	7.62ns	1.76*	20.00 ns	175.80 ns	30.40 ns	4.55 ns
		±0.06	±0.12	±0.04	±1.25	±2.15	±1.16	±0.06
	10	1.66ns	7.76ns	2.40*	28.42*	182.53 ns	38.78 ns	6.75 ns
		±0.07	±0.14	±0.10	±1.11	±2.27	±2.40	±0.01
	25	2.80*	7.88ns	4.55**	32.80*	210.27*	56.72*	9.44*
		±0.04	±0.11	±0.10	±2.13	±2.45	±2.13	±1.04
	50	3.24*	8.08*	6.99**	42.50*	235.89*	72.90*	13.77*
		±0.07	±0.10	±0.14	±2.10	±3.27	±2.31	±1.02
	75	3.96*	8.38*	8.82**	50.34**	245.94**	92.39*	15.75*
		±0.07	±0.12	±0.13	±2.55	±3.58	±2.77	±1.02
	100	4.23**	8.67*	10.40**	54.30**	256.40**	122.90**	17.40*
		±0.05	±0.19	±0.10	±2.30	±3.10	±3.20	±1.07
Summer	0 (Control)	0.47	7.62	0.53	19.25	165.55	26.88	2.59

	±0.05	±0.12	±0.01	±1.10	±2.62	±1.05	±0.07
5	1.35ns	7.65ns	1.80*	21.60ns	180.90 ns	33.45 ns	4.68 ns
	±0.04	±0.17	±0.05	±1.26	±2.75	±1.09	±0.07
10	1.78ns	7.81ns	2.48*	30.87*	197.30 ns	41.90 ns	7.24 ns
	±0.08	±0.13	±0.10	±1.70	±2.88	±2.10	±1.06
25	2.86*	7.92ns	4.78**	34.50*	216.70*	59.50*	10.58*
	±0.05	±0.19	±0.12	±2.96	±2.98	±2.42	±1.02
50	3.34*	8.15*	7.25**	44.75*	248.90*	76.55*	15.88*
	±0.09	±0.15	±0.11	±2.44	±3.44	±2.75	±1.05
75	4.02*	8.45*	8.86**	53.90**	253.67**	98.45*	18.57*
	±0.09	±0.18	±0.14	±2.80	±3.66	±2.44	±1.06
100	4.31**	8.74*	11.00**	55.86**	262.50**	125.60**	20.05*
	±0.06	±0.13	±0.16	±2.66	±3.24	±3.75	±1.09

Note: Mean ± SE of three values; ns, *, ** non-significant or significant at $P < 0.05$ or $P < 0.01$.

Table 2b. Effects of Sewage Sludge (SS) Concentration and Season Interaction on Physico-Chemical Characteristics of Soil after Cultivation of *S. Melongena* in Both Seasons

Season × %SS		TKN (mg kg^{-1})	PO_4^{3-} (mg kg^{-1})	SO_4^{2-} (mg kg^{-1})	Fe (mg kg^{-1})	Cd (mg kg^{-1})	Cr (mg kg^{-1})	Cu (mg kg^{-1})	Mn (mg kg^{-1})	Zn (mg kg^{-1})
Rainy	0	25.12	2.45	77.80	2.84	0.54	0.34	2.40	0.51	2.80
	(Control)	±1.00	±0.11	±2.10	±0.08	±0.06	±0.02	±0.07	±0.04	±0.08
	5	32.50 ns	6.45 ns	84.33 ns	2.95*	1.10*	1.06*	3.30*	1.20*	3.12*
		±1.01	±0.13	±2.02	±0.10	±0.04	±0.03	±0.06	±0.05	±0.10
	10	38.40**	10.75 ns	96.40 ns	4.28*	1.75*	1.27*	4.25*	1.60*	4.56*
		±1.02	±0.15	±2.09	±0.11	±0.05	±0.05	±1.05	±0.07	±0.13
	25	53.20**	15.80*	124.32*	6.78**	2.25**	2.49**	6.75**	2.18**	6.88**
		±1.09	±0.23	±2.14	±0.17	±0.03	±0.03	±1.04	±0.03	±0.10
	50	72.65**	24.20**	146.73*	8.95**	4.02**	2.80**	8.80**	3.78**	9.89**
		±1.05	±0.25	±2.15	±0.16	±0.08	±0.05	±1.03	±0.09	±0.12
	75	82.80**	39.57**	164.83**	10.88**	5.06**	3.47**	10.98**	4.50**	12.98**
		±1.06	±0.27	±1.98	±0.13	±0.05	±0.04	±1.00	±0.05	±0. 08
	100	117.59**	54.65**	178.64**	12.95**	5.74**	3.72**	12.98**	5.98**	16.80**
		±1.04	±0.18	±1.60	±1.00	±0.03	±0.07	±1.02	±0.07	±0.05
Summer	0	26.10	2.48	78.10	2.90	0.55	0.35	2.42	0.52	2.81
	(Control)	±1.01	±0.10	±2.00	±0.12	±0.04	±0.03	±0.08	±0.02	±0.06
	5	34.58ns	7.23 ns	88.20 ns	2.10*	1.18*	1.13*	3.35*	1.27*	3.22*

	±1.02	±0.22	±2.11	±0.16	±0.08	±0.04	±0.06	±0.01	±0.08
10	55.80**	12.66*	102.77*	4.43*	1.86*	1.32*	4.36*	1.72*	4.77*
	±1.04	±0.20	±2.19	±0.17	±0.06	±0.05	±1.03	±0.03	±0.09
25	60.70**	18.23*	129.66*	7.45**	2.45**	2.52**	6.89**	2.45**	6.89**
	±1.00	±0.13	±2.23	±1.02	±0.05	±0.08	±1.08	±0.04	±0.10
50	78.40**	28.58**	155.40*	9.80**	4.28**	2.87**	8.96**	3.88**	10.66**
	±1.07	±0.18	±2.10	±1.08	±0.04	±0.09	±1.03	±0.08	±0.11
75	87.34**	43.97**	172.80**	11.78**	5.34**	3.54**	11.23**	4.77**	13.25**
	±1.06	±0.19	±2.15	±1.03	±0.07	±0.03	±1.08	±0.06	±0.10
100	122.50**	59.67**	183.90**	14.95**	5.78**	3.78**	13.30**	6.04**	17.24**
	±1.04	±0.17	±2.11	±1.05	±0.07	±0.05	±1.10	±0.03	±0.12

Note: Mean ±SE of three values; ns, *** non-significant or significant at $P < 0.01$.

During the present study, ANOVA indicated that seasons, sewage sludge concentrations and the their interaction showed significant ($P < 0.05$/$P < 0.01$/$P < 0.001$) effect on EC, pH, OC, Na^+, K^+, Ca^{2+}, Mg^{2+}, TKN, PO_4^{3-} , SO_4^{2-} , Cd, Cr, Cu, Fe, Mn and Zn of the soil (Table 2). Additionally, EC, OC, Na^+, K^+, Ca^{2+}, Mg^{2+}, Fe^{2+}, TKN, PO_4^{3-}, SO_4^{2-} Zn, Cd, Cu, Mn and Cr were found to be positively correlated with sewage sludge concentration in both seasons (Table 3). The use 100% dose of sewage sludge increased the OC (10.40 and 11.00 mg kg^{-1}), Na^+ (54.30 and 55.86 mg kg^{-1}), K^+ (256.40 and 262.50 mg kg^{-1}), Ca^{2+} (122.90 and 125.60 mg kg^{-1}), Mg^{2+} (17.40 and 20.50 mg kg^{-1}), TKN (117.59 and 122.50 mg kg^{-1}), PO_4^{3-} (54.65 and 59.67 mg kg^{-1}), SO_4^{2-} (178.64 and 183.90 mg kg^{-1}), Fe^{2+} (12.95 and 14.95 mg kg^{-1}), Cd (5.74 and 5.78 mg kg^{-1}), Cr (3.72 and 3.78 mg kg^{-1}), Cu (12.98 and 13.30 mg kg^{-1}), Mn (5.98 and 6.04 mg kg^{-1}) and Zn (16.80 and 17.24 mg kg^{-1}) in both seasons. The findings are in accordance with Alcantara et al. (2009) who also reported the higher concentration of Cu (5.39 mg kg^{-1}), Cr (0.62 mg kg^{-1}), Fe (51.53 mg kg^{-1}), Mn (5.54 mg kg^{-1}), Ni (2.22 mg kg^{-1}) and Zn (30.27 mg kg^{-1}) in an Oxisol treated with domestic and industrial sewage sludge. Kidd et al. (2007) also reported the considerable increase in OC, TKN, P, Cd, Cu, Mn and Zn in agricultural soil amended by long term application of sewage sludge.

Table 3. Coefficient of Correlation (r) between Sewage Sludge (SS) and Soil Characteristics in both Seasons

Sewage sludge/soil characteristics	Season	r-value
Sewage sludge versus soil EC	Rainy	+0.95
	Summer	+0.97
Sewage sludge versus soil pH	Rainy	+0.80
	Summer	+0.84

Sewage sludge versus soil OC	Rainy	+0.92
	Summer	+0.94
Sewage sludge versus soil Na^+	Rainy	+0.93
	Summer	+0.94
Sewage sludge versus soil K^+	Rainy	+0.90
	Summer	+0.92
Sewage sludge versus soil Ca^{2+}	Rainy	+0.89
	Summer	+0.90
Sewage sludge versus soil Mg^{2+}	Rainy	+0.90
	Summer	+0.92
Sewage sludge versus soil TKN	Rainy	+0.98
	Summer	+0.98
Sewage sludge versus soil PO_4^{3-}	Rainy	+0.82
	Summer	+0.83
Sewage sludge versus soil SO_4^{2-}	Rainy	+0.80
	Summer	+0.84
Sewage sludge versus soil Fe^{2+}	Rainy	+0.96
	Summer	+0.97
Sewage sludge versus soil Cd	Rainy	+0.94
	Summer	+0.96
Sewage sludge versus soil Cr	Rainy	+0.92
	Summer	+0.95
Sewage sludge versus soil Cu	Rainy	+0.97
	Summer	+0.98
Sewage sludge versus soil Mn	Rainy	+0.90
	Summer	+0.92
Sewage sludge versus soil Zn	Rainy	+0.97
	Summer	+0.98

The present study observed that contamination factor (Cf) of the metals indicated that Mn (11.72 and 11.61) was highest while Cu (5.49 and 5.40) was lowest in both seasons after amendment with 100% sewage sludge. The Cf of metals were in the order of Mn > Cr > Cd > Zn > Cu after treatment with sewage sludge in both seasons (Figure 1). Kumar and Chopra (2012) also reported the most contamination factor for Cd (10.75 and 11.60) and least for Cu (5.47 and 6.52) in distillery effluent fertigated soil.

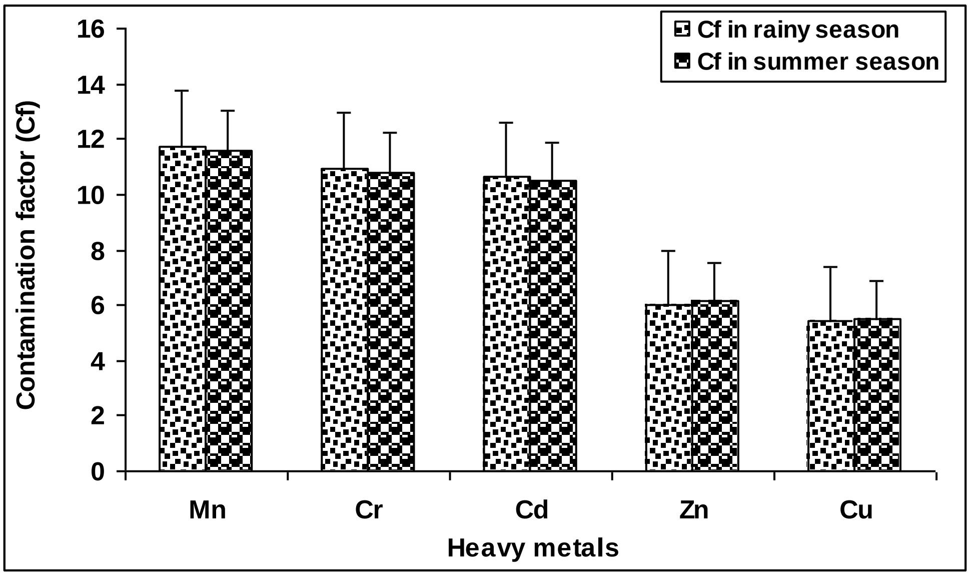

Figure 1. Contamination Factor of Heavy Metals in Soil after Sewage Sludge Treatment in both Seasons

Note: Error bars are standard error of the mean.

The results indicated that application of sewage sludge considerably increased the nutrients Na^{+}, K^{+}, Ca^{2+}, Mg^{2+}, Fe^{2+}, OC, TKN, PO_4^{3-}, SO_4^{2-} and heavy metals Zn, Cd, Cu, Mn and Cr in the soil. This is likely due to the presence of significant quantity of these chemical species in the sewage sludge (Table 2). Although, the contents of heavy metals were found to be below the maximum levels permitted for Cd (6.0 mg kg^{-1}), Cr (10.0 mg kg^{-1}), Cu (270 mg kg^{-1}) and Zn (600 mg kg^{-1}) for soil in India (BIS, 2010). However, there was gradual build up of these metals in the sewage sludge amended soil and this is likely due to the addition of more organic matter in the soil which provides more binding sites for heavy metals. Therefore, the application of sewage sludge significantly increased the nutrients as well as metals content in the soil.

3.3 Effect of Sewage Sludge on Seed Germination of S. Melongena

The percent seed germination of *S. melongena* after amendment with sewage sludge is shown in Figure 2. During the present study, most seed germination (90.00 and 88.00%) of *S. melongena* was observed with 50%, while that of the least seed germination (80.00%) was noted with 100% treatment of sewage sludge (Figure 2). The seed germination of *S. melongena* was negatively correlated ($r = -0.34$ and -0.33) with sewage sludge doses in both seasons. At germination stage, ANOVA indicated that seasons showed insignificant ($P > 0.05$) effect on seed germination of *S. melongena* while sewage sludge concentrations and their interaction with seasons showed significant ($P < 0.05$) effect on seed germination of *S. melongena*. The findings in agreement with Dursan et al. (2005) who reported that the germination of pepper (*Capsicum annuum* L.) was decreased with the increase in the concentration of sewage sludge. In the present study, the higher concentrations (50% to 100%) of sewage sludge did not support seed germination. Kumar and Chopra (2013b) also reported that the germination of French bean (*Phaseolus vulgaris* L.) was decreased when the concentration of sewage sludge increased. The higher concentrations of sewage sludge lowered germination of *S. melongena*. It may be likely due to the presence of more contents of salts and heavy metals in the sewage sludge at higher concentrations of sewage sludge.

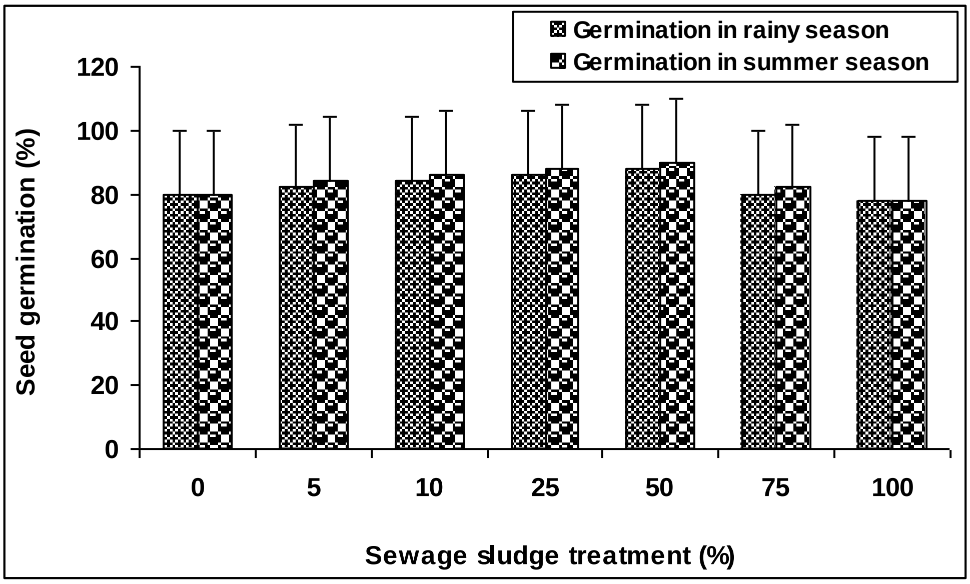

Figure 2. Seed Germination of *S. Melongena* after Sewage Sludge Treatment in both Seasons

Note: Error bars are standard error of the mean.

3.4 Effect of Sewage Sludge on Vegetative Growth of S. Melongena

In the present study, at vegetative growth at 75 days the maximum plant height (90.36 and 95.60 cm), root length (14.55 and 15.23 cm), dry weight (280.95 and 294.40 g), chlorophyll content (0.854 and 0.862mg./g.f.wt) and LAI/plant (3.19 and 3.21) of *S. melongena* were observed with 50% concentration of sewage sludge in both seasons. The minimum plant height (60.20 and 62.80 cm), root length (10.00 and 10.25 cm), dry weight (210.40 and 220.60 g), chlorophyll content (0.526 and 0530 mg./g.f.wt) and LAI/plant (3.00 and 3.05) of *S. melongena* were observed with control while moderate plant height (75.25 and 80.60 cm), root length (10.24 and 12.50 cm), dry weight (250.30 and 264.80 g), chlorophyll content (0.654 and 0.667 mg./g.f.wt) and LAI/plant (3.10 and 3.15) of *S. melongena* were noted with 100% sewage sludge in both seasons.

During the present study, plant height, root length, dry weight, chlorophyll content and LAI/plant of *S. melongena* were positively correlated with sewage sludge concentrations in both seasons (Table 4). Prabhu et al. (2009) also reported that the growth of *S. melongena* was recorded to be positively correlated with different concentrations of sewage sludge. The sewage sludge concentrations showed significant ($P < 0.05$) effect on plant height, chlorophyll content and LAI of *S. melongena*. Seasons showed insignificant (P > 0.05) effect on root length, dry weight of *S. melongena*. The interaction of seasons and sewage sludge concentrations only affected plant height, chlorophyll content and LAI of *S. melongena*. Likewise, Kumar and Chopra (2013b) reported that the maximum vegetative growth attributes like plant height, root length, dry weight, chlorophyll content and leaf area index of French bean (*Phaseolus vulgaris* L.) was noted with 40% concentration of sewage sludge. Furthermore, the growth of *P. vulgaris* was decreased with the increase in sewage sludge concentration from 60% to 100%. The findings were also supported by Fukuoka et al. (2010) who reported that the growth of eggplant was decreased due to the application of more concentration of sewage effluent.

Table 4. Coefficient of Correlation (r) between Sewage Sludge and *S. Melongena* in both Seasons

Sewage sludge/*S. melongena*	Season	r - value
Sewage sludge versus plant height	Rainy	+0.65
	Summer	+0.68
Sewage sludge versus root length	Rainy	+0.44
	Summer	+0.47
Sewage sludge versus dry weight	Rainy	+0.50
	Summer	+0.52
Sewage sludge versus chlorophyll content	Rainy	+0.75
	Summer	+0.76
Sewage sludge versus LAI	Rainy	+0.80
	Summer	+0.82
Sewage sludge versus no. of leaves/plant	Rainy	+0.50
	Summer	+0.50
Sewage sludge versus leaves length	Rainy	+0.48
	Summer	+0.48
Sewage sludge versus crop yield/plant	Rainy	+0.47
	Summer	+0.49
Sewage sludge versus Cd	Rainy	+0.95
	Summer	+0.96
Sewage sludge versus Cr	Rainy	+0.94
	Summer	+0.95
Sewage sludge versus Cu	Rainy	+0.97
	Summer	+0.98
Sewage sludge versus Mn	Rainy	+0.84
	Summer	+0.86
Sewage sludge versus Zn	Rainy	+0.97

	Summer	+0.98
Sewage sludge versus total carbohydrate	Rainy	+0.75
	Summer	+0.76
Sewage sludge versus crude protein	Rainy	+0.70
	Summer	+0.72
Sewage sludge versus dietary fiber	Rainy	+0.84
	Summer	+0.86

Vegetative growth of *S. melongena* was decreased at higher concentrations i.e., 75% to 100% of sewage sludge. It might be due to that more content of heavy metals in the higher concentrations of sewage sludge, which lowered the plant height, root length, dry weight, chlorophyll content and LAI/plant of *S. melongena*. Vegetative growth is related with the development of new shoots, leaves and leaf area (Khan et al., 2007; Kumar & Chopra, 2012). Plant height, root length, dry weight and LAI/plant of *S. melongena* were noted higher at 50% of sewage sludge, and it may be likely due to maximum uptake of nitrogen, phosphorus and potassium by plants. The improvement of vegetative growth may be attributed to the role of potassium in nutrient and sugar translocation in plants and turgor pressure in plant cells (Khan et al., 2007). It is also involved in cell enlargement and in triggering young tissue or mersitematic growth (Porra, 2002; Khan et al., 2007; Kumar & Chopra, 2012). Chlorophyll content was found higher due to the use of 50% sewage sludge in both seasons, and is likely due to Fe, Mg and Mn contents in the sewage sludge, which are associated with chlorophyll synthesis (Yadav et al., 2002; Zhou et al., 2012). Thus, 50% concentration of sewage sludge contains optimum contents of nutrients required for maximum vegetative growth of *S. melongena*.

3.5 Effect of Sewage Sludge on Maturity of S. Melongena

At maturity stage (110 days after sowing), the maximum number of leaves/plants (25.00 and 28.00), leaves length (12.20 cm and 12.50 cm) yield/plant (1250.25 and 1290.40 g) at I harvest, (1450.30 and 1520.56 g) at II harvest and (1120.40 and 1190. 50 g) at III harvest of *S. melongena* were recorded with 50% concentration of sewage sludge in both seasons. The numbers of leaves/plant, leaves length, crop yield/plant and HI of *S. melongena* were positively correlated with all concentrations of sewage sludge in both seasons (Table 4). Numbers of leaves/plant, leaves length, crop yield/plant and HI of *S. melongena* were not affected by seasons, sewage sludge concentrations and their interaction. The least number of leaves/plants (18.00 and 20.00), leaves length (10.12 cm and 10.30 cm) yield/plant (980 and 995 g) at I harvest, (1020.10 and 1050.60 g) at II harvest and (1010.25 and 1018.40 g) at III harvest of *S. melongena* were with the control while modest leaves/plants (22.00 and 23.00), leaves length (11.10 cm and 11.25 cm) yield/plant (1060.20 and 1080.35 g) at I harvest, (1170.47 and 1186.20 g) at II harvest and (1135.40 g and 1145.30 g) at III harvest of *S. melongena* were recorded with 100% sewage sludge in both seasons. At maturity stage the 50% sewage sludge favored crop yield of *S. melongena*. This is likely due to the presence of optimal contents of K, Fe, Mg and Mn in 50% sewage sludge;

higher concentrations i.e., 75% to 100% of sewage sludge lowered crop yield of *S. melongena*, thus inhibited the growth of *S. melongena* and it may be due to the presence of more heavy metals, which reduced the uptake of nutrients.

The role of K, Fe, Mg and Mn at maturity is important and associated with synthesis of chlorophyll, and enhances the crop yield (Saxena & Diwakar, 2012; Kumar & Chopra, 2012). The K, Fe, Mg and Mn contents could enhance the yield of eggplants (*S. melongena*) as reported by Fukuoka et al. (2010), Kumar and Chopra (2013b) reported that the maximum crop yield of French bean (*Phaseolus vulgaris* L.) was noted with 40% concentration of sewage sludge. Moreover, the crop yield of *P. vulgaris* was decreased with the increase in sewage sludge concentration from 60% to 100%.

3.6 Effect on Heavy Metals and Biochemical Components in S. Melongena

The contents of various heavy metals and biochemical components *viz.*, total carbohydrate, crude protein and dietary fiber were positively correlated with all concentrations of sewage sludge in both seasons (Table 4). Seasons, all concentrations of sewage sludge and their interaction affected all the metals and biochemical components in *S. melongena*. The 25% to 100% concentrations of sewage sludge showed significant ($P < 0.05$/$P < 0.01$) effect on Cd, Cr, Cu, Mn and Zn, total carbohydrate, crude protein and dietary fiber in *S. melongena.* The most contents of Cd, Cr, Cu, Mn and Zn in *S. melongena* were observed with 100% sewage sludge (Figures 3, 4). It might be due to the presence of more heavy metals in 100% sewage sludge, which added more metals in the soil environment. The accumulation of Cr, Cu, Mn and Zn except Cd in *S. melongena* was noted below the permissible limit of FAO/WHO standards for Cd (0.20 mg Kg^{-1}), Cr (2.30 mg Kg^{-1}), Cu (40.00 mg Kg^{-1}) and Zn (60.00 mg Kg^{-1}) (FAO/WHO, 2011). The contamination factor (Cf) of heavy metals was affected in both seasons. The Cf of various metals was in the order of Cr > Cd > Cu > Mn > Zn in *S. melongena* after application of sewage sludge (Figure 5). The highest contamination factor was noted for Cr (35.42 and 32.25), while the least was found for Zn (10.85 and 10.82) in *S. melongena* with 100% sewage sludge in both seasons. Interestingly, five metals examined in the present study have different chemical properties and as a result each metal has peculiar accumulation. The diverse accumulation of metals might be due to the number of electrons in the d-levels of the atom. Metals with completely filled d orbitals such as (d10, Cr and Zn) may be least incorporated into the *S. melongena* plants due to lower reactivity and more stability imparted by the completely filled d orbitals. The lower reactivity and stability of these metals reduce the rate of various reactions such as absorption, ionic exchange, redox reactions, precipitation and dissolution through which plants take heavy metals from soils (Chopra et al., 2013; Kumar & Chopra, 2015).

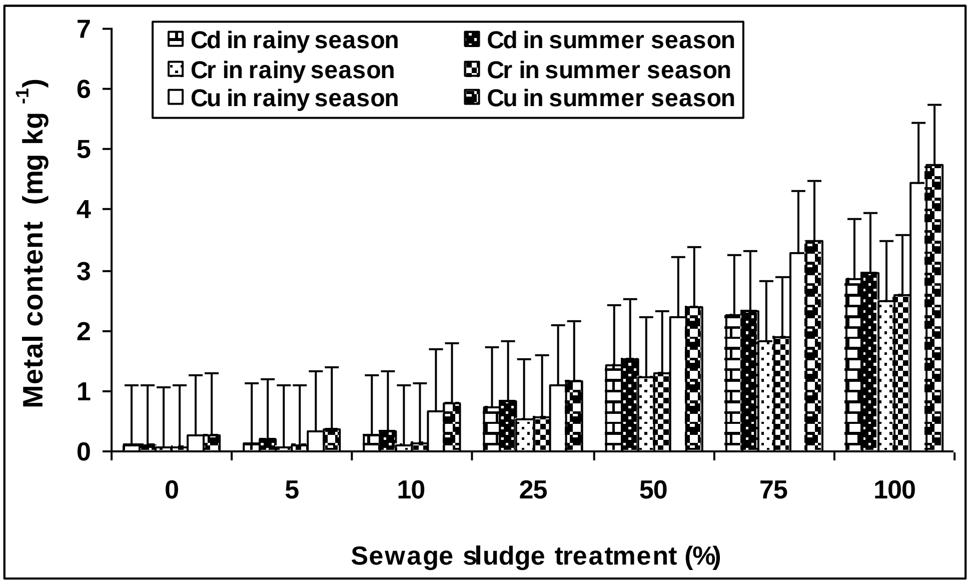

Figure 3. Content of Cd, Cr and Cu in *S. Melongena* after Sewage Sludge Treatment in both Seasons

Note: Error bars are standard error of the mean.

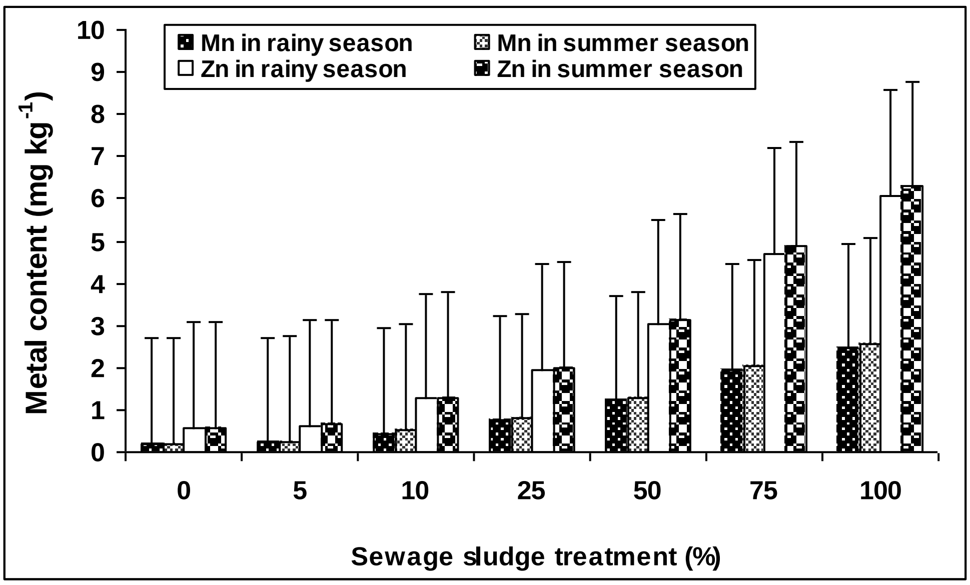

Figure 4. Content of Mn and Zn in *S. Melongena* after Sewage Sludge Treatment in both Seasons

Note: Error bars are standard error of the mean.

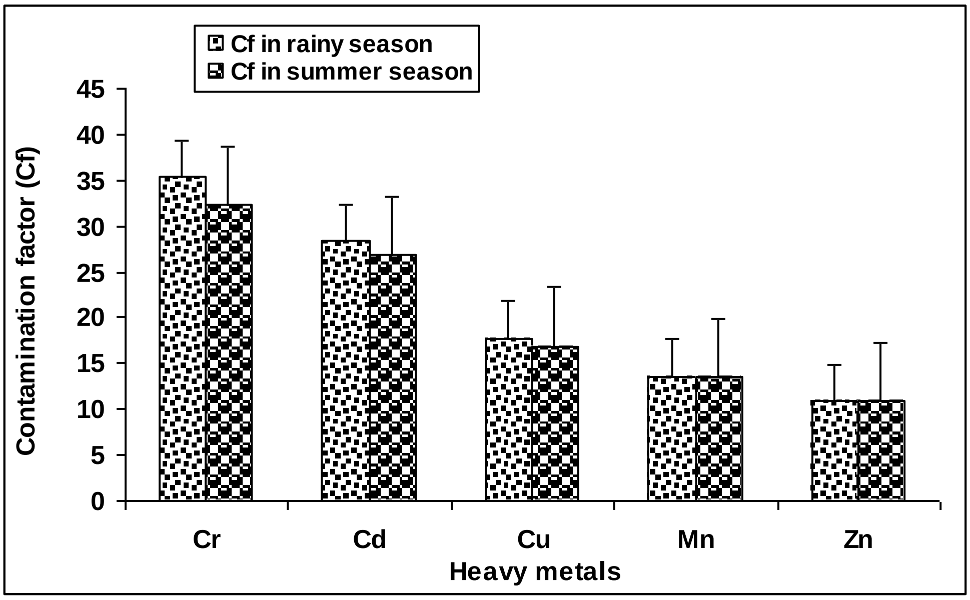

Figure 5. Contamination Factor of Heavy Metals in *S. Melongena* after Sewage Sludge Treatment in both Seasons

Note: Error bars are standard error of the mean.

The results showed that the contents of these metals were more at 75% to 100% concentrations of sewage sludge, and likely inhibited growth of *S. melongena*. The 50% concentration of sewage sludge favored vegetative growth, flowering and maturity of *S. melongena*. This is likely due to optimal uptake of these metals by crop plants, which supports various biochemical and physiological processes. The results are in conformity of Kumar and Chopra (2013b) who reported the accumulation of metals in the order of Fe > Zn > Cd > Cu > Cr > Pb after amended with sewage sludge. Nunome et al. (2001) also reported the contamination of Cd, Cu, Cr, Zn and Pb in soil and *S. melongena* after application of sewage sludge and effluent. During the present study, maximum contents of total carbohydrate, crude protein and dietary fiber were noted with 50% sewage sludge (Figures 6-8). The contents of total carbohydrate, crude protein and dietary fiber were decreased from 75% to 100% doses of sewage sludge. The findings are in agreement with Kumar and Chopra (2014c; 2015) who reported that the contents of total carbohydrate, crude protein and crude fiber in French bean (*Phaseolus vulgaris* L.) and spinach (*Spinacia oleracea* L.) were decreased with the increase in the concentration of sugar mill and paper mill effluent, respectively when these effluents were used for fertigation.

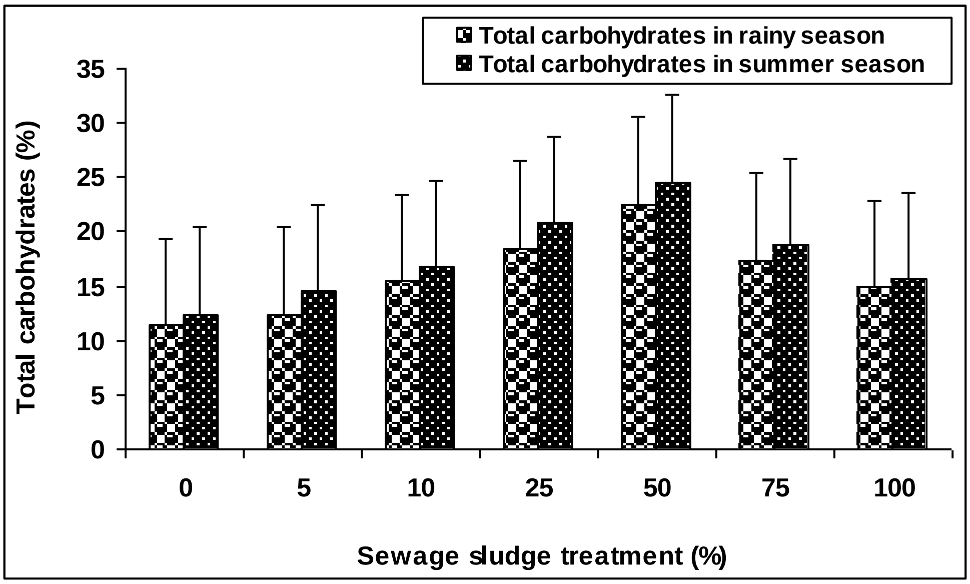

Figure 6. Total Carbohydrates in *S. Melongena* after Sewage Sludge Treatment in both Seasons

Note: Error bars are standard error of the mean.

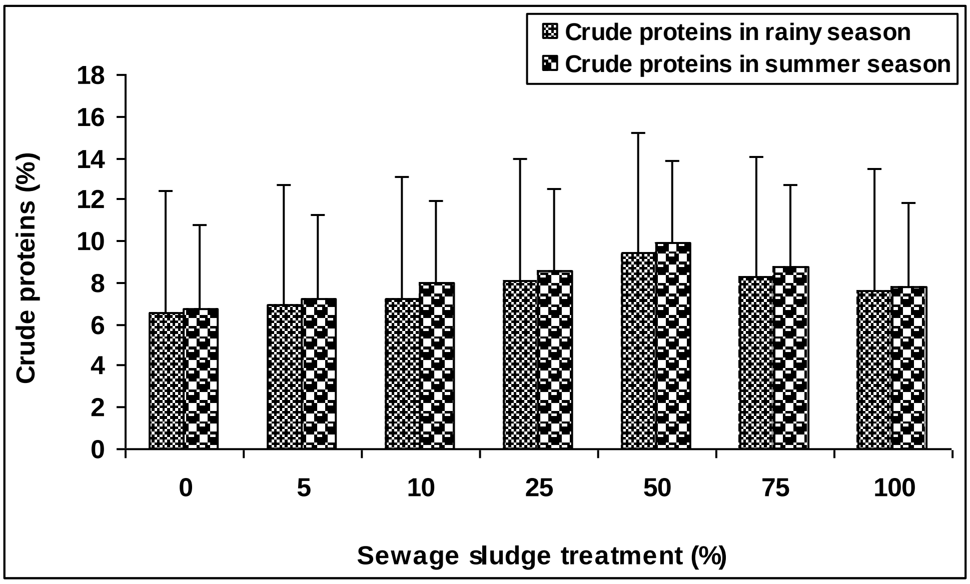

Figure 7. Crude Proteins in *S. Melongena* after Sewage Sludge Treatment in both Seasons

Note: Error bars are standard error of the mean.

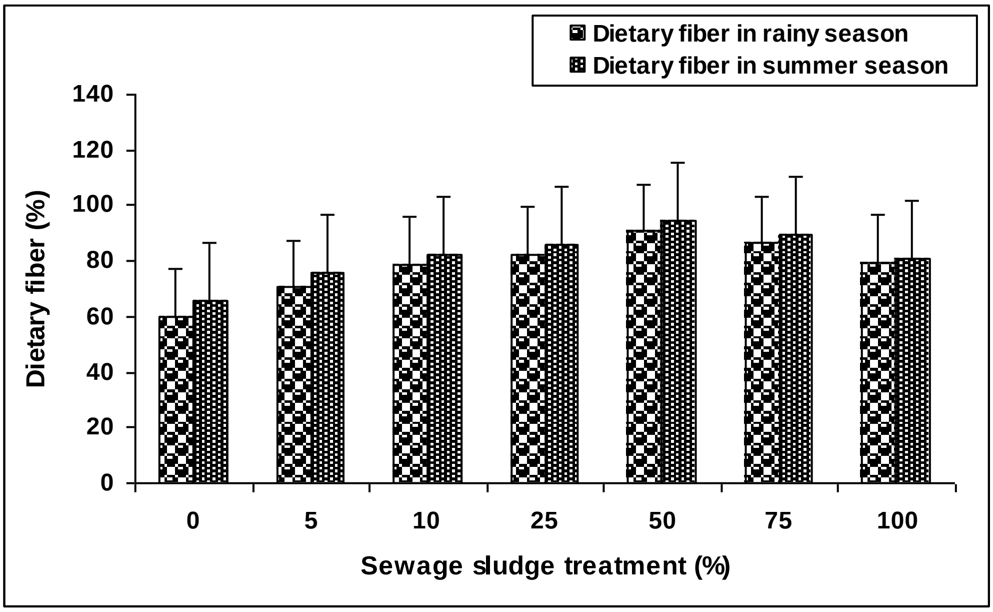

Figure 8. Dietary Fiber in *S. Melongena* after Sewage Sludge Treatment in both Seasons

Note: Error bars are standard error of the mean.

4. Conclusions

This study concluded that sewage sludge was a source of nutrients and heavy metals. The sewage sludge amended soil, lead to accumulation of heavy metals in *S. melongena*. The use of sewage sludge increased the contents of nutrients and heavy metal in the soil in both seasons. The most agronomic growth of *S. melongena* was observed with 50% doses of sewage sludge in both seasons. The growth of *S. melongena* was inhibited at higher doses (75% to 100%) and it might be due to the presence of more contents of heavy metals at these doses. The accumulation of Cd, Cu, Cr, Mn and Zn was increased in soil and *S. melongena* from 5% to 100% concentrations of sewage sludge in both seasons. The contamination factor (Cf) of various heavy metals was in order of Mn > Cr > Cd > Zn > Cu for soil and Cr > Cd > Cu > Mn > Zn for *S. melongena* plants after application with sewage sludge. The contents of Cu, Mn and Zn except Cd and Cr in *S. melongena* were noted under the permissible limit of FAO/WHO standards. Thus, this study recommends that there may not be a significant threat posed by heavy metals contamination of *S. melongena* and soils. Taking into consideration the long-term applications of sewage sludge compost would carry a risk of progressive of heavy metals to toxic levels. Thus, use of sewage sludge to the soil and plant analysis is needed to check the levels of sewage sludge borne metals used as soil amendments. Therefore, sewage sludge can be governed as organic manure for kitchen or urban gardens in lower proportion to avoid the contamination of soil and *S. melongena*.

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