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Effects of sewage sludge amendment on heavy metal accumulation and consequent responses of *Beta vulgaris* plants

R.P. Singh, M. Agrawal *

Ecology Research Laboratory, Department of Botany, Banaras Hindu University, Varanasi 221 005, India

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Abstract

Use of sewage sludge, a biological residue produced from sewage treatment processes in agriculture is an alternative disposal technique of waste. To study the usefulness of sewage sludge amendment for palak (*Beta vulgaris* var. Allgreen H-1), a leafy vegetable and consequent heavy metal contamination, a pot experiment was conducted by mixing sewage sludge at 20% and 40% (w/w) amendment ratios to the agricultural soil. Soil pH decreased whereas electrical conductance, organic carbon, total N, available P and exchangeable Na, K and Ca increased in soil amended with sewage sludge in comparison to unamended soil. Sewage sludge amendment led to significant increase in Pb, Cr, Cd, Cu, Zn and Ni concentrations of soil. Cd concentration in soil was found above the Indian permissible limit in soil at both the amendment ratios.

The increased concentration of heavy metals in soil due to sewage sludge amendment led to increases in heavy metal uptake and shoot and root concentrations of Ni, Cd, Cu, Cr, Pb and Zn in plants as compared to those grown on unamended soil. Accumulation was more in roots than shoots for most of the heavy metals. Concentrations of Cd, Ni and Zn were more than the permissible limits of Indian standard in the edible portion of palak grown on different sewage sludge amendments ratios. Sewage sludge amendment in soil decreased root length, leaf area and root biomass of palak at both the amendment ratios, whereas shoot biomass and yield decreased significantly at 40% sludge amendment. Rate of photosynthesis, stomatal conductance and chlorophyll content decreased whereas lipid peroxidation, peroxidase activity and protein and proline contents, increased in plants grown in sewage sludge-amended soil as compared to those grown in unamended soil.

The study clearly shows that increase in heavy metal concentration in foliage of plants grown in sewage sludge-amended soil caused unfavorable changes in physiological and biochemical characteristics of plants leading to reductions in morphological characteristics, biomass accumulation and yield. The study concludes that sewage sludge amendment in soil for growing palak may not be a good option due to risk of contamination of Cd, Ni and Zn and also due to lowering of yield at higher mixing ratio.

Keywords: Sewage sludge; Heavy metals accumulation; Biomass; Physiological characteristics; Yield

1. Introduction

Use of sewage sludge in agriculture is a worldwide practice, and is a very effective sludge disposal technique. Sewage sludge enables the recycling of valuable components such as organic matter and many plant nutrients (Logan and Harrison, 1995). The macronutrients in sewage sludge such as N, P, K, etc. serve as good source of plant nutrients, but very rarely the urban sewage system transport only domestic sewage to the treatment plants. Industrial as well as storm water runoff from the nearby area are frequently discharged into the sewage system. As the population is increasing, so the loading of sewage sludge is also increasing for disposal.

Sewage sludge composition is variable and may contain high levels of toxic metals (Smith, 1992), which limit their land application due to food chain contamination (Chaney, 1990). While other nutrients present in sewage sludge are subjected to leaching or removed through crop uptake or

* Corresponding author. Tel.: +91 542 2368156; fax: +91 542 2368174.
E-mail address: madhoo58@yahoo.com (M. Agrawal).

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runoff, heavy metals may persist in soil for a much longer time and may result in phytotoxicity. High concentrations of heavy metals such as Zn, Ni, Cd and Cu were reported in soil amended with sewage sludge (Hernandez et al., 1991). Heavy metal accumulation in plant tissues was also reported in plants grown on sludge-amended soil (Moreno et al., 1997). 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).

Sludge amendment (80, 130 and 160 t ha^{-1}) increased the average dry weight of sunflower plants (*Helianthus annus* L.) as 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). Tsakou et al. (2001) also found faster development and significant root and shoot biomass production of *Gossypium hirsutum* grown on sludge-amended soil. In contrast, Moreno et al. (1997) reported negative effects of sludge amendment on yield of *Lactuca sativa*.

The fertility benefits from sewage sludge amendment can be achieved against the potential hazards of heavy metal contamination by screening the plants sensitivity at different sludge amendment ratios. Plants differ in their heavy metal uptake, accumulation and tolerance levels. In view of the above, the present study was carried out to assess the effect of different sewage sludge amendment ratios on physiological characteristics and yield of palak (*Beta vulgaris* L. var. Allgreen H-1). Heavy metal accumulation in soil and different parts of plants was also quantified to find out the correlation between heavy metal accumulation and plant response. Palak is a nutritious leafy vegetable commonly grown in suburban areas having frequent sewage sludge uses.

2. Materials and methods

2.1. Study area

The experiment was conducted at the agricultural farm, Banaras Hindu University, a suburban area of Varanasi, located in the eastern Gangetic plain of the Indian subcontinent at 25°14′N latitude, 82°3′E longitude and 76.19 m above the sea level. The experiment was carried out between April and May 2004. This period of the year is characterized by mean monthly maximum temperatures between 37.6 and 41.1 °C and mean monthly minimum temperatures between 21.4 and 25.8 °C. The total rainfall was 3.13 mm. Maximum relative humidity varied from 47% to 66% and minimum from 16% to 29%.

2.2. Experimental design and raising of plants

The experiment was carried out in earthen pots of 30 cm diameter and 30 cm depth. There were three treatments i.e. unamended soil (control), 20% sewage sludge (w/w) and 40% (w/w) sewage sludge amendments. Sewage sludge was collected from Sewage Treatment Plant (STP), Dinapur, Varanasi, which has water treatment capacity of 80 MLD covering the entire Varanasi city except Banaras Hindu University (BHU) and Diesel Locomotive Works (DLW).

Soil from the field of agricultural farm was dug out upto the depth of 30 cm, left in the field for air-drying and then mixed properly. Sewage sludge was also air dried and then grounded properly to mix uniformly. Three heaps of soil was prepared. One was used as unamended control, whereas in remaining two, 20% and 40% (w/w) sewage sludge amendments were done. The sewage sludge was mixed uniformly with soil and left for 10 days in the field. Before filling the pots of respective treatments, separate heaps were again mixed properly. Fifteen pots each were prepared for different treatments. Pots were filled uniformly upto the height of 26 cm and left for five days. Three replicates each from different treatments were collected for soil physico-chemical analyses after fifteen days of initial mixing. Necessary moisture level was maintained and thereafter six seeds of palak were sown manually at equivalent distances in each pot.

Palak (*Beta vulgaris* L. var. Allgreen H-1), a cheap and popular green vegetable consumed mainly as an iron source in the diet was chosen for the experimental work. Indian Agricultural Research Institute (IARI), New Delhi developed this variety. It is a very high yielding late bolting variety.

After germination, plants were thinned to three in each pot. The pots were kept in open field condition to provide identical light and temperature conditions to all the treatments. Identical water regime was maintained for all the treatments through out the growth period of plants.

2.3. Soil analyses

Soil samples collected in triplicate from control, 20% and 40% sewage sludge amendment were air dried, crushed, passed through a sieve of 2 mm mesh size and then stored separately for further analyses. For analysis of heavy metals in soil samples, 1 g air-dried sample was digested in 20 ml of tri acid mixture (HNO_3:H_2SO_4:$HClO_4$: 5:1:1) for 8 h at 80 °C following the method described by Allen et al. (1986). After complete digestion solution was filtered and the filtrate was analyzed separately for heavy metals using Atomic Absorption Spectrophotometer (Model 2130, Perkins Elmer, Inc., Norwalk, CT, USA).

The pH of the soil at different treatments was measured in the suspension of 1:5 (w/v) with the help of pH meter (Model EA940, Orion, USA) standardized with pH 4, 7 and 9.2 reference buffers. Conductivity was measured by conductivity meter (Model 303, Systronics, India). Na^+, K^+ and Ca^{+2} were extracted from soil in ammonium acetate solution following repeated leaching procedure and then concentrations were determined with the help of