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Biosorption of lead from aqueous solutions of varied pH by kale plants (*Brasicca oleraceae var acephala*)

V. Emongor, University of Botswana, vemongor@bca.bw

Abstract

This study was undertaken to evaluate the effect of irrigating kale plants (*Brassica oleraceae var acephala*) with water of variable pH on lead uptake and partitioning. Lead was applied as lead nitrate (300 mg/kg). Kale plants were irrigated with water having pH values of 4.5, 5.5, 6.5, 7.1 (tap water, used as control), 7.5 and 8.5, respectively, throughout the experimental period. The results showed that lead accumulated more in kale roots, followed by the stem, leaf blade and leaf petiole. Irrigating kale plants with acidic water (pH 4.5-6.5) increased lead accumulation in kale roots, stem, and leaf petioles and blades. As the pH of the irrigation water became more alkaline (pH 7.1-8.5), there was a significant ($P<0.0001$) decrease in lead accumulation in kale leaf petiole, leaf blade, stem and roots. The results also showed that irrigating kale plants with water of pH 6.5 increased kale leaf size, fresh leaf yield and dry matter accumulation. It was concluded that lead uptake was significantly ($P<0.0001$) increased when the medium of growth pH was acidic (pH 4.5-6.5). Therefore, to minimize lead uptake and optimize growth and fresh leaf yield of kale plants, the pH of the medium should be between 6.5 and 7.0.

Keywords: Kale, lead, uptake, pH, partitioning, plant tissue accumulation.

Introduction

One of the major pathways of adult and children exposure to contaminants such as heavy metals and organic compounds is through the food chain (IPCS, 1989; WHO, 1990). Barzi et al., (1996) reported that in Australia, contamination of vegetables in urban soils occurred through plant uptake of contaminants when grown in contaminated soil and/or surface contamination through aerial deposition. Vegetables and fruits get contaminated as a result of some of the following activities: use of industrial sludge as residential landfill; proximity of horticultural farms to mines or smelters and motorways (in countries that still use petrol containing lead); use of banned insecticides for crop production (arsenic/DDT-some soils contaminated with DDT and lead arsenate); agricultural lands contaminated with Pb-based paints; and use of municipal sewage effluent and sludge for irrigation and organic soil amendment, respectively. As a toxic substance flows up the food chain, each organism experiences more concentrated levels of toxicity than the organism below it (Florence et al., 1996; Emongor et al., 2005).

Pollution of the environment with toxic metals and radioactive waste has dramatically increased since the beginning of the industrial revolution (Emongor et al., 2005). The heavy metals of widespread concern to human health are mercury, cadmium, lead, arsenic, chromium, copper, and zinc (Mason, 1996; Lazrova et al., 2005). Nriagu (1988) estimated that over one billion human beings are currently exposed to elevated concentrations of toxic metals and metalloids in the environment and several million people may be suffering from subclinical metal poisoning. Heavy metals suppress the immune system, leading to increased susceptibility to disease in animals. Some of the heavy metals are carcinogenic (Peakall, 1992). Some heavy metals such as lead are neurotoxic and children are particularly vulnerable because of the rapidly developing nervous system (Cohen, 2005). Lead is known to inhibit the activity of three critical enzymes (5-aminolaevulinate dehyratase (ALA-D), coproporphyrinogen oxidase (COPRO-O) and ferrochelatase (FERRO-C)) critical in haem synthesis, causing abnormal concentrations of haem precursors in blood and urine (IPCS, 1995).

Lead (Pb) is discharged into the environment from automobile exhausts (Foth, 1990), industrial sources (Griesmer, 2001) and hunting, recreataional shooting and fishing (Kapsch et al., 1996), and it eventually reaches the soil. The soil is the most important repository in terrestrial ecosyetems for contaminants of anthropogenic origin (Nriagu & Pacyna, 1988; Nriagu, 1989). The Pb content of soils is influenced by anthropogenic activities and long-and short-range airborne transport of Pb from various sources. In the soil, Pb is converted to forms unavailable to plants (Foth, 1990). Any lead that is absorbed by plants tends to remain in the roots and is not transported to the shoots (Foth, 1990). Foth (1990) further reported that soil must become much polluted with Pb before significant amounts move into the tops of the plants (Foth, 1990). Interest in the soil as a source of Pb for crop plants has been increased by the concern over airborne Pb from automobile exhausts. Just how much Pb is deposited directly on the leaf surface and soil, and later taken up by plants is not known. Lead in the soil may be relatively insoluble (as a sulphate, carbonate or oxide), soluble, adsorbed onto clays, adsorbed and coprecipitated with sesquioxides, adsorbed onto colloidal organic matter, or complexed with organic moieties in soil (U.S. EPA, 1986; IPCS, 1989). However, the behaviour of Pb in the soil suggests that much of the lead in food crops comes from atmospheric contamination (Brady, 1984).

Soil pH greatly influences the availability of both nutrients and toxins for uptake by plant roots (Brady & Weil, 2002; IPCS, 1989). Lead has been measured in super phosphate fertilizer at concentrations as high as 92 mg/kg (IPCS, 1995). Sewage sludge, used as a source of nutrients in agriculture may contain up to 1000 mg/kg Pb (IPCS, 1995). Soils receiving heavy sewage sludge and municipal sewage effluent applications may accumulate large quantities of Pb and other heavy metals and organic contamitants. Soil pH affects the mobility of pollutants in the soil by influencing the rate of biochemical breakdown, solubility and adsorption to colloids. Thus, soil pH is a critical factor in predicting the likelihood that a given pollutant will contaminate groundwater, surface water and food chain (Brandy & Weil, 2002; IPCS, 1989). It is very important to know the factors that influence the availability of heavy metals and other organic contaminants in the soil especially where municipal effluents (particularly if contaminated with industrial wastewater discharge is suspected) are used for irrigation. Kale was used in the study because it is one of the major leafy vegetables eaten and grown in Africa. In urban and peri-urban areas of some cities in Africa, leafy vegetables such as kale, amaranth, cowpeas and spinach beet are irrigated with municipal sewage effluent and fertilized with treated sewage sludge, which may be contaminated with heavy metals. The objective of this study was to evaluate the effect of irrigating kale plants (*Brassica oleraceae var acephala*) with water of variable pH on Pb uptake and partitioning.

Materials and Methods

Study site

The study was carried out in a net greenhouse in The Botswana College of Agriculture (BCA), Sebele farm (24˚33'S, 25˚54E, and 994 m above sea level) during the periods of January to June 2006. The climate in Sebele is semi arid with an average annual rainfall (30 year mean) of 538 mm. The soils are shallow, ferruginous tropical soils, mainly consisting of medium to coarse grain sands and sandy loams with a low water holding capacity and subject to crusting after heavy rain. The soil are deficient in phosphorus, have low levels of mineral nitrogen and low organic matter (Emongor et al., 2004). The soil that was used for the experiment was sandy loam.

Soil sampling.

The soil was excavated from the BCA-garden using shovel, mixed several times into one large heap. The excavation was done to an average depth of 30 cm from one continuous area in order to minimize variability. The soil was sampled into 90 medium size black polyethylene bags by filling the bags with 10 kg soil. The soil was analyzed in the laboratory for pH, cation exchange capacity, calcium, sodium, potassium, magnesium and organic carbon. The soil used for the study had low sodium content of 0.09 meq/100g, potassium of 0.39 meq/100g, magnesium of 0.16 meq/100g and calcium content was 9.53 meq/100g). The organic carbon content was 1.04 meq/100g, CEC content of 7.53 meq/100g, and pH of 6.5. Seeds of *Brasicca oleraceae var acephala* (kale) were planted in polysterene trays, which were transplanted into polyethylene bags four weeks after emergence.

Experimental design

The treatments comprised of a control (tap water (pH 7.1), water of pH 4.5, 5.5, 6.5, 7.5 and 8.5. The experimental design used was a completely randomized design with three replicates. Three hundred (300 mg/kg) milligrams per kilogram of lead as lead nitrate was top-dressed to all kale plants three weeks after transplanting. After lead nitrate application, the plants were irrigated with water of adjusted pH (4.5, 5.5, 6.5, 7.1, 7.5 and 8.5). The plants were watered three times per week with water of adjusted pH throughout the experimental period. At each watering one litre of water was applied (3 litres of water per week per plant). Nitric acid was used to lower the pH of water (treatments that had the pH lower than 6.5). Treatments with pH above 7, sodium hydroxide was used to raise the water pH. The spacing between the bags was 30 cm and 60 cm between rows.

Variables determined

The dependent variables that were determined included; leaf petiole and blade length, leaf dry matter and water content, stem root, leaf petiole and blade lead contents. Fresh leaf yield was determined by harvesting plant leaves from three selected plants in each replicate. Leaf harvests were done every two weeks for a period of 12 weeks. The samples were harvested into brown paper bags and were weighed in the laboratory using Mettler PM 3000 balance to determine the leaf fresh weight. The leaf petiole length and blade length were measured using a 30 cm ruler at every leaf harvest. After measuring the leaf petiole and blade lengths, the leaf petioles and blades were separated and put in already weighed brown paper bags. The stems and roots of three selected kale plants per replicate were put in brown paper bags.

The leaf petiole and blade, stem and root samples were dried at 66°C to constant weight for 72 hours. The leaf water content was determined by subtracting the dry matter from their corresponding fresh weight. The percentage leaf dry matter and water content were calculated. Dried samples were ground and 1.25g of composite sample were digested in 98% sulphuric acid and 30% selenium powder in a digestion block at 330°C for 8 hours (AOAC, 1995). The lead in the leaf petioles and blades, roots and stems were determined using Atomic Absorption Spectrometry (AA-6650 Shimadzu).

Data analysis

Data collected was subjected to analysis of variance using the Statistical Analysis System (SAS, 2005). Treatment means were separated using the Least Significant Difference (LSD) at P=0.05.

Results

The results from the study showed that kale plants have the ability to take up Pb and partition it to different plant parts (Figure 1). The results also showed that out of 300 ppm of Pb applied, kale plants absorbed a maximum of about 1.03% Pb (Figure 1). The uptake of Pb by kale plants depended on the medium pH (Figure 1). Kale plants irrigated with alkaline water (pH 7.1 to 8.5) had significantly lower Pb uptake (Figure 1). As the irrigation water pH increased from acidity to alkalinity Pb uptake by kale plants significantly decreased (Figure 1). Kale plants irrigated with water having a pH of 4.5 had the highest Pb accumulation (Figure 1). Irrespective of the pH of the irrigation water, the roots had significantly ($P<0.0001$) higher Pb accumulation compared to stem, leaf blade and petiole (Figure 1). Kale leaf petiole accumulated the least Pb compared to roots, stem and leaf blade (Figure 1). The kale roots had 2x more lead than the leaf petiole and blade, respectively (Figure 1). The Pb content in the various plant organs decreased in the following order: roots> stem>leaf blade>leaf petiole (Figure 1).

Irrigating kale plants with water of pH 4.5 and 5.5 had no significant effect on leaf blade and petiole lengths, respectively (Figure 2). Kale plants irrigated with water of pH 6.5 resulted in plants with longer petiole and leaf blade length than plants irrigated with water of pH 7.1, 7.5 and 8.5 (Figure 2).

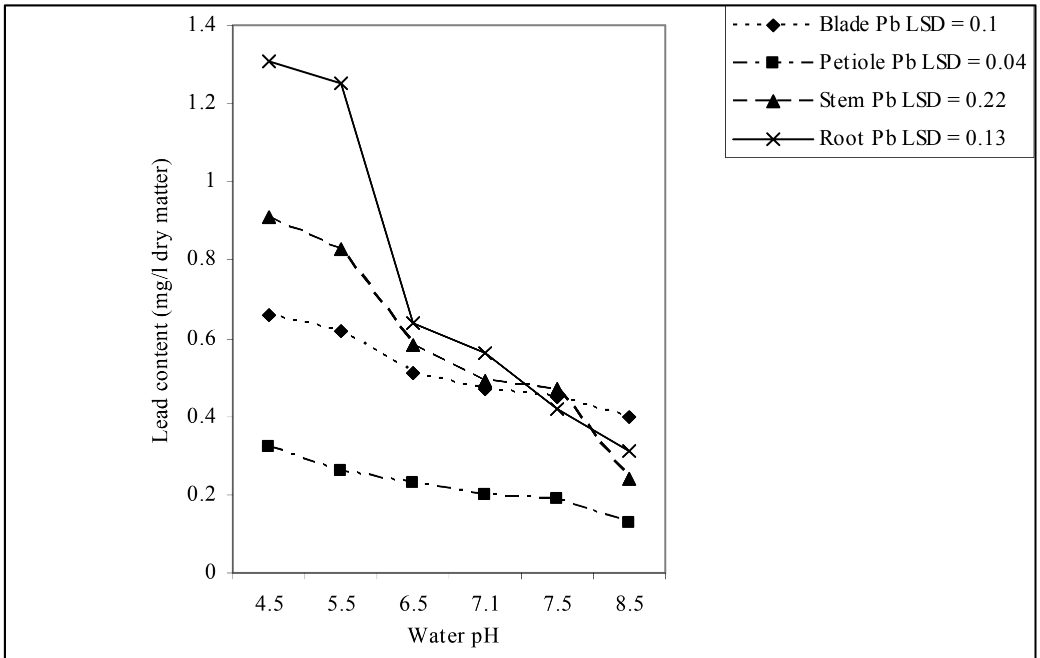

Figure 1. Effect of irrigating kale plants on Pb partitioning

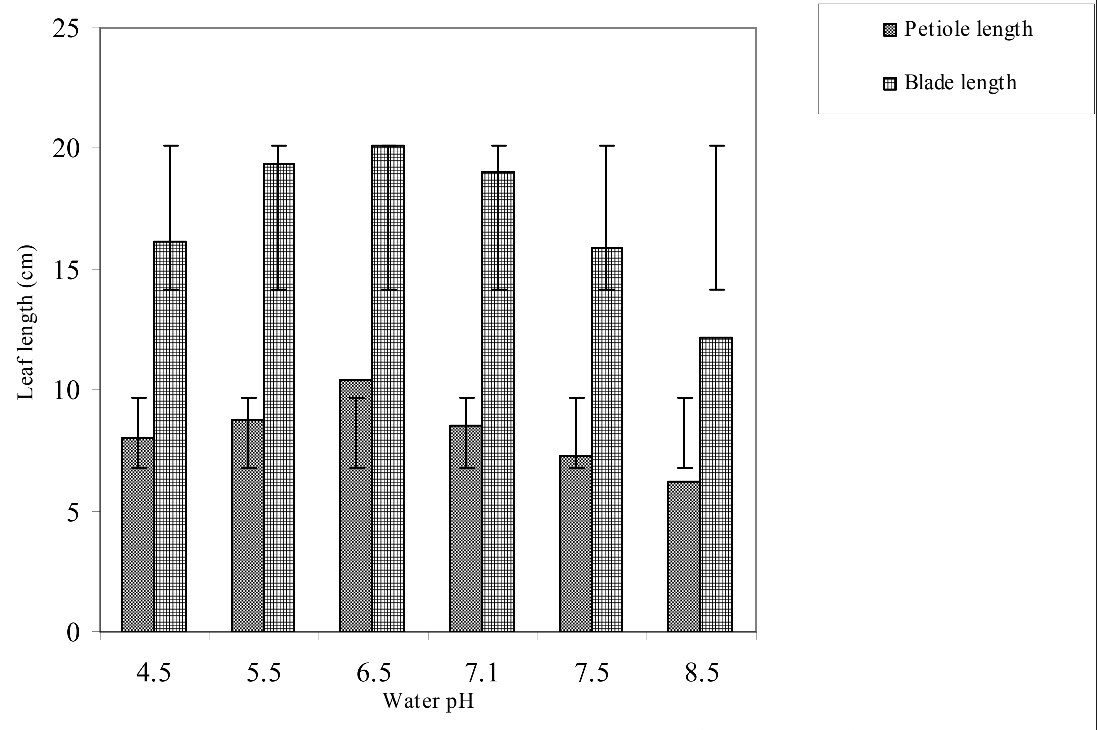

Figure 2. Effect of irrigating kale plants with water of adjusted pH on leaf size

There was a significant decrease in leaf petiole and blade length of plants irrigated with water of pH 7.1, 7.5 and 8.5 (Figure 2). Irrigating kale plants with water of pH 8.5 resulted in plants with the shortest leaf petiole and leaf blade lengths compared with plants irrigated with water having pH of 6.5 (Figure 2).

Kale plants irrigated with water of pH 6.5 had significantly (P<0.001) higher dry matter and lower

water content than plants irrigated with water of pH 4.5, 5.5, 7.1, 7.5 and 8.5, respectively (Figure 3). While kale plants irrigated with water of pH 4.5, 5.5, 7.1, 7.5 and 8.5, did not differ significantly in their dry matter and water contents, respectively (Figure 3). In general as acidity and alkalinity in the irrigation water increased kale leaf dry matter decreased and leaf water content increased, respectively (Figure 3).

Irrigating kale plants with water of pH 6.5 significantly ($P<0.001$) increased the fresh leaf yield compared to plants irrigated with water of pH 4.5, 5.5, 7.1, 7.5 and 8.5 (Figure 4). There was no significant difference in fresh leaf yield between irrigated kale plants with water of pH 4.5, 5.5, 7.1 and 7.5 (Figure 4). However, there was a significant ($P<0.005$) decrease in fresh leaf yield of kale plants irrigated with water of pH 8.5 compared to those plants irrigated with water having pH of 5.5, 6.5, 7.1, and 7.5 (Figure 4). There was also a significant reduction in fresh leaf yield of plants irrigated with water of pH 4.5 compared to irrigated with water of pH 6.5 (Figure 4).

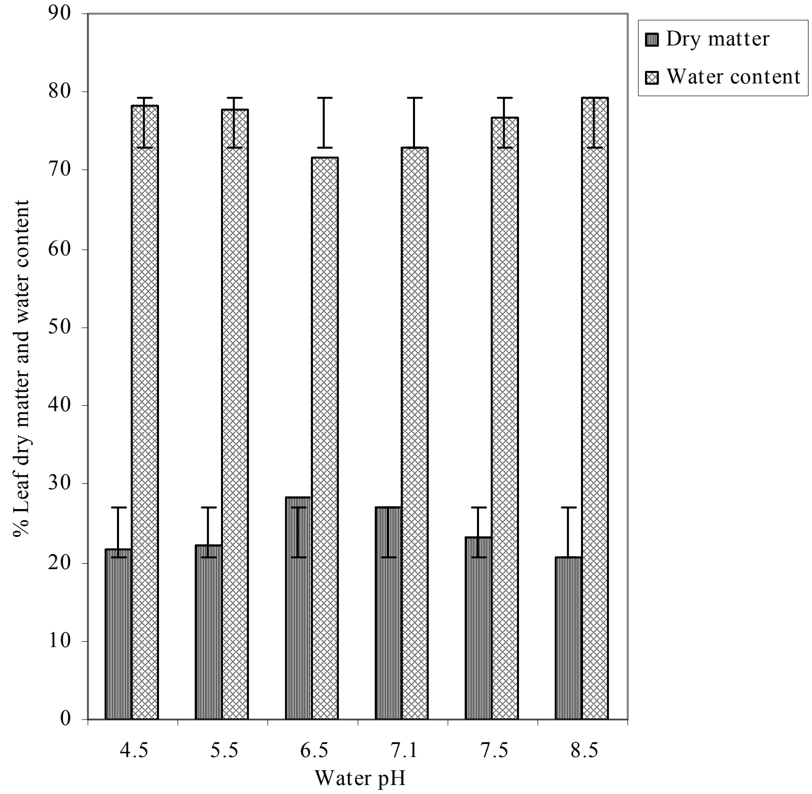

Figure 3. Effect of irrigating kale plants with water of adjusted pH on leaf dry matter and water content

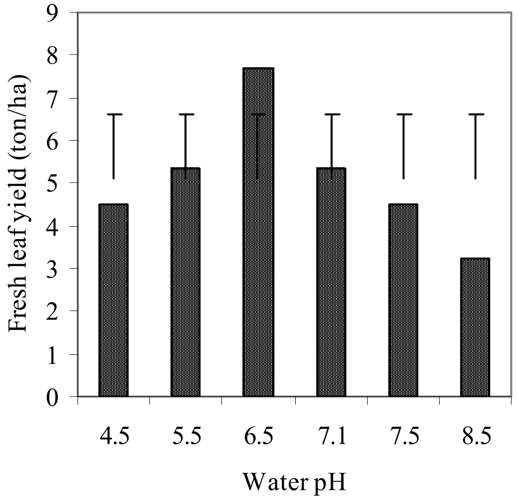

Figure 4. Effect of irrigating kale plants with water of adjusted pH on yield

Discussion

Generally kale plants absorbed Pb regardless of the soil pH level. Kale plants irrigated with acidic water (pH 4.5, 5.5 and 6.5) had significantly ($P<0.001$) higher lead accumulation in roots, stems and leaf petioles and blades compared to plants irrigated with alkaline water (pH 7.1, 7.5 and 8.5). The results also showed that as the irrigation water pH increased from 4.5 to 8.5, the Pb uptake and accumulation decreased significantly. The dynamic equilibrium between metals in solution and soil-solid phase is determined by the properties of the soil and composition of the soil solution. This equilibrium in turn controls the availability of heavy metals and contaminants for plants for uptake. The soil factors that control the equilibrium are soil pH, ionic strength, and presence of ligands in soil solution that may affect sorption, soil organic matter and dissolved organic material (Oliver & Naidu, 2003; Helmke & Naidu, 1996; Jones & Jarvis, 1981). Soil pH, content of humic and fulvic acids, and the amount of organic matter has been reported to influence the content and mobility of Pb in soils (IPCS, 1995). The increased solubility of Pb due to the acidic conditions (pH 4.5-7.1) in the growth medium in the current study plus the low organic matter content (1.04meq/100g) may have attributed to Pb uptake by kale plants. Organic-Pb complexes become more soluble and bioavailable within soil pH range of 4 to 6 (U.S. EPA, 1986). Foth (1990) and Brady (1984) reported that Pb was less available to plants when the soil pH was above 6.5 and high in phosphorus and organic matter. Davies et al. (1987) reported that there was a positive linear relationship between Pb concentrations in plants and soil. The results of the current study have shown that bioavailable Pb to plants is highly pH dependent. The results of the current study also suggests that in order to minimize Pb uptake by kale plants, the medium of growth and irrigation water pH should be higher than 6.5. The results from the current study also revealed that Pb accumulated more in the roots compared to the stem or leaves of kale plants. This suggests that root vegetables may have high Pb uptake and accumulation, which may be a potential source of Pb poisoning depending on the soil pH and Pb content. The results of the current study are in agreement with the findings of Foth (1990), who reported that Pb absorbed by plants tends to remain in the roots. However, the results of the current study are in disagreement with the findings that Pb absorbed by plants is not transported to the shoots (Foth, 1990) because the kale plants translocated Pb to leaf blade and petiole and stem (shoot).

The leaf blade had more Pb content than the leaf petiole due to the larger surface area of the leaf blade compared to the leaf petiole. The higher Pb content in the leaf blade compared to the leaf petiole also suggests possible atmospheric Pb deposition. Mushak et al. (1989) reported that production sources of Pb included root vegetables, root Pb uptake from the soils or atmospheric Pb deposition into leafy vegetables. Valley (2003) reported that in urban gardens, Pb was more concentrated in the roots of plants, though some Pb was detected in the shoots. The present study further showed that irrespective of irrigation water pH, kale plants absorbed Pb and partitioned it to different plant parts. However, more Pb was retained in the roots than the shoot. This is of great concern to vegetables grown in urban or peri-urban areas, where the levels of Pb are relatively high in the air, soil and water due to pollution from the industries (paints, car batteries) and leaded gasoline. In order to safe guard the health of the urban population, governments have to put policies to regulate the mount of Pb in paints, car batteries, public drinking water and phase out leaded gasoline.

Soil pH affects the availability of soil nutrients to plants (Brady, 1984). Kale plants irrigated with water of pH 6.5 significantly increased kale fresh leaf yield and dry matter accumulation compared to plants irrigated with water having a pH of 4.5, 5.5, 7.1, 7.5 and 8.5. From the results of the current study, pH 6.5 seemed to be the best pH for kale growth and development because the plants had higher fresh leaf yield and larger leaf size than plants irrigated with water of pH 4.5, 5.5, 7.1, 7.5 or 8.5. Which suggested that at pH of 6.5 plant nutrients were available to kale plants for growth and development. Soils with pH 7.5 have been reported to cause iron, manganese, copper, zinc and boron to be less available to plants (Brady, 1984). The availability of Ca and P decreases at pH values above 6.0. The low availability of P at pH> 6.0 may explain why there was Pb uptake at pH> 7.0 in the current study.

In conclusion irrigating kale plants with water of acidic pH significantly increased the uptake of Pb compared to alkaline water. Amending acidic soils contaminated with Pb with agricultural lime (calcium carbonate) to pH greater than 7 may minimize Pb ingestion by man and animals. Kale plants also partitioned Pb differently among plant parts with more Pb in the roots, followed by the stem and leaves, respectively. It was also concluded that the optimum pH for kale growth is pH 6.5 because it resulted in kale plants with higher fresh yield and dry matter and relatively low Pb content. It was recommended that due to the uptake of Pb by kale plants at all pH values under study, the governments of various countries should formulate policies that regulates Pb content in public drinking and irrigation water, paint, gasoline, water pipes and all industrial emissions which are the possible Pb pollution sources.

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