The Influence of Lead Hyperaccumulators on the Uptake of Lead by Vegetables

Laura Knapp

Senior, Department of Civil Engineering
University of Nebraska-Lincoln
Omaha, NE 68182-0178
lknapp@unomaha.edu

Jodi Sangster

Graduate Student, Department of Civil Engineering
University of Nebraska-Lincoln
Omaha, NE 68182-0178
jsangster@unomaha.edu

Shannon L. Bartelt-Hunt

Associate Professor, Department of Civil Engineering
University of Nebraska-Lincoln
Omaha, NE 68182-0178
sbartelt2@unl.edu

Abstract-Lead is a persistent soil pollutant present in many urban environments. In 2003, a 27 square mile region in Omaha NE became the newest addition to the National Priorities List for lead contamination. This large area encompasses 15,000 residential properties as well as an urban community garden. Previous work performed at the urban garden detected the presence of lead in produce. Research has shown that vegetation grown in these contaminated soils can bioaccumulate lead and become a pathway for exposure in humans. Although the relationship between soil concentration and plant uptake is unknown, it has been demonstrated that plants exhibit differential uptake. This study focused on the possible role of metal-hyperaccumulating plants to minimize the uptake of lead in adjacent vegetation. To conduct this study, soil contaminated with lead was used to grow tomatoes, carrots, and amaranth alongside sunflowers and mustard greens, the hyperaccumulators of interest. Samples of both the soils and produce were taken in triplicate and processed with nitric acid. Lead concentrations were then determined using atomic adsorption spectroscopy. The resulting data showed higher concentrations of lead in the hyperaccumulators and suggested that the presence of the hyperaccumulators may have had an impact on the amount of lead available to surrounding vegetation. Further research may provide additional tools in reducing human exposure to lead from produce grown in urban gardens.

Index Terms – Lead, soil, vegetable, uptake

INTRODUCTION

Lead is a toxic heavy metal commonly found in urban environments^{1,2}. It accumulates in the body and can have a negative impact on the gastrointestinal tract, kidneys, and central nervous system. In children, these effects may be fatal¹. Various activities including the once large-scale practices of using lead-based paint and burning leaded gasoline have introduced lead

contaminants into the environment where it has become bound to the soil matrix and is able to persist for years¹⁻³. Although average lead concentrations in agricultural soils range from less than 1 mg/kg to 135 mg/kg³, lead soil concentrations exceeding 1000 mg/kg are commonly reported in urban areas^{2,4}. Recent research is showing that plant tissue can bioaccumulate lead when grown in contaminated soils⁴⁻⁸ and that the lead contained in this plants tissue is also bioavailable to humans as evidenced by elevated blood lead levels in children in urban communities with a prevalent culture of backyard gardening and reliance on homegrown produce⁴. In 2003, a 27 square mile region in Omaha NE, containing 15,000 residential properties as well as the City Sprouts community garden, was added to the National Priorities List for lead contamination. Previous testing of the community garden does show lead contamination in both ground soil and raised bed soil⁵. Although the concentrations of lead are well below the Environmental Protection Agency's (EPA) action limit of 400 mg/kg, testing has proved its presence in the produce grown on site. The United States Department of Agriculture (USDA) and Food and Drug Administration (FDA) do not currently have standards regulating the quality of soil used to grow produce⁹.

Conventional methods of soil remediation are costly and invasive¹⁰, thus raised beds gardening is becoming an increasingly popular option to reduce contact between produce and the underlying contaminated soil^{4,9}. These beds, however, are subject to contamination from windblown particles originating in neighboring contaminated yards. They are therefore not a permanent method of reducing lead exposure in urban gardens⁴. Another, though less conventional, method of soil remediation is phytoremediation¹⁰. This option, which uses metal-accumulating plants to remove heavy metals from soil over long periods of time, does not provide complete removal of lead from contaminated soils¹⁰. However, the use of competitive phytoremediation as a means of controlling uptake of lead by surrounding vegetation rather than as a means to remove contaminants from soil has not been previously considered. This would provide the added benefits of managing produce lead levels while reducing remediation time and maintaining a productive garden during the process.

The biggest challenge with the use of competitive phytoremediation is the limited information available regarding the influence of plant type on lead uptake. Research indicates leafy plants such as spinach, lettuce, and amaranthus may contain between 3.3 to 24 mg/kg on a dry weight basis⁶⁻⁸ and arugula, mustard, and collards may contain between 13 and 24 mg/kg on a wet weight basis^{4,5} depending on washing techniques. Additionally, fruiting plants such as tomatoes, eggplant, and okra have been found to contain lead concentrations up to 3.7 mg/kg on a wet weight basis⁵ while known metal hyperaccumulators such as sunflowers may contain on average 360 mg/kg in root biomass and 47 mg/kg in leaf material⁴.

The aim of this study is to investigate the ability of lead hyperaccumulating plants to minimize the uptake of lead into surrounding vegetation. The goal is to provide the City Sprouts community garden and the backyard gardening community with additional and inexpensive tools for reducing lead exposure in humans while continuing to have productive use of their backyard gardens. This study was conducted as an independent undergraduate research project.

METHODS AND MATERIALS

Sample Collection and Preparation:

To better understand the extent to which hyperaccumulators are able to uptake lead and make it unavailable to surrounding vegetation, the relationship between the uptake of each

hyperaccumulator with various vegetables was evaluated. Sunflowers and mustard greens were chosen as the hyperaccumulators of interest due to their high uptake rates. Carrots, amaranth, and tomatoes were chosen to be representative of commonly grown vegetables where the edible portion are roots, shoots, and fruits, respectively. Lead contaminated soil was obtained through the EPA office responsible for the cleanup of properties located within the Omaha, Nebraska Lead Superfund site. The US EPA office provided lead contaminated soils at two concentrations: a 'low' lead concentration of 350 mg/kg and a 'high' lead concentration of 450 mg/kg. These concentrations were found to be representative of those commonly found in backyard gardens.

Triplicates of the carrots, amaranth, and tomatoes were planted in the contaminated soil in three groups: a positive control group without any hyperaccumulator, an experimental group planted alongside sunflowers, and an additional experimental group planted alongside mustard greens. In early spring, the carrots, amaranth, sunflowers, and mustard greens were started from seed in the contaminated soil while the tomatoes were started as young plants in the contaminated soil to replicate common practices of backyard gardening. All plants were grown in the greenhouse on the campus of the University of Nebraska at Omaha and were watered and fertilized as needed. After a 6 week growing period, samples of the edible portion of each specimen were collected from each experimental group. These samples were thoroughly washed and chopped. Soil samples were taken from each group at the close of the growing season. All samples were dried in a drying oven before proceeding with the lead analysis.

Total Lead Extraction Procedure:

Lead from the soil and vegetable samples were extracted using a nitric acid/concentrated hydrogen peroxide process. Briefly, approximately 1 gram of sample was boiled in concentrated nitric acid to dissolve organic material and lead. This was allowed to cool before the addition of hydrogen peroxide. The remaining extract was concentrated to approximately 25mL of solution. Soil samples were centrifuged to remove any remaining particulates. Vegetable samples were filtered using a glass fiber filter to remove particulates. This extraction procedure (USEPA SW-846, Method 3050) was used in a previous publication by the authors⁵.

Measurement of Total Lead Content:

The liquid extracts were analyzed for total lead using atomic absorption spectroscopy (AAS). This process involves igniting the liquid extract while exposing it to a specific wavelength of light. The amount of light that passes through the sample can be recorded and compared to that for solutions containing known concentrations of lead using a standard curve. For lead, there are several wavelengths of light that can be used, the most common being 283.3 nm. This wavelength was used for the soil extracts. However, the vegetables contained significantly less lead than the sediments. To allow for more sensitive measurements of lead, it was necessary to use 217 nm for these samples. This analysis procedure was used previously by the authors⁵.

RESULTS AND DISCUSSION

Average values for the concentrations found in each experimental group can be seen in the following tables. Concentrations expressed in Tables 1-4 are on a wet weight basis.

TABLE 1
LEAD CONCENTRATIONS IN TOMATO WITH AND WITHOUT
HYPERACCUMULATORS, LOW LEAD LEVELS

	<u>Soil Concentration</u> (mg/kg)	<u>Concentration in Tomato</u> (mg/kg)	<u>Hyperaccumulator</u> <u>Concentration (mg/kg)</u>
Tomato			
No hyperaccumulator control	286	0.3	
Tomato with Sunflowers	346	0.7	14.6
Tomato with Mustard Greens	697	0.1	10.9

TABLE 2
LEAD CONCENTRATIONS IN TOMATO WITH AND WITHOUT
HYPERACCUMULATORS, HIGH LEAD LEVELS

	<u>Soil Concentration</u> (mg/kg)	<u>Concentration in Tomato</u> (mg/kg)	<u>Hyperaccumulator</u> <u>Concentration (mg/kg)</u>
Tomato			
No hyperaccumulator control	1734	3	
	1084	0	
Average:	1409 ± 459	1.5 ± 2	
Tomato with Sunflowers	1150	1	0
	1281	0	29
Average:	1215 ± 93	0.5 ± 0.7	14.5 ± 20.7
Tomato with Mustard Greens	1304	3	30
	1467	0	0
Average:	1385 ± 115	1.5 ± 2.1	15 ± 21

TABLE 3
LEAD CONCENTRATIONS IN CARROT WITH AND WITHOUT
HYPERACCUMULATORS, LOW LEAD CONCENTRATIONS

	<u>Soil Concentration</u> (mg/kg)	<u>Concentration in Carrot</u> (mg/kg)	<u>Hyperaccumulator</u> <u>Concentration (mg/kg)</u>
Carrots			
No hyperaccumulator control	254	1.6	
	204	1.8	
Average:	229±35.4	1.7±0.14	
Carrot with Sunflower	360	2.2	8.4
	327	2.3	0.0
	287	1.3	8.8
Average:	325±36.6	1.9±0.55	5.7±4.97
Carrot with Mustard Greens	~346	2.7	3.6
	346	0.7	8.6
	~346	1.3	
Average:	346±0	1.6±1.03	6.1±3.54

TABLE 4
LEAD CONCENTRATIONS IN AMARANTH WITH AND WITHOUT
HYPERACCUMULATORS, LOW LEAD CONCENTRATIONS

	<u>Soil Concentration</u> (mg/kg)	<u>Vegetable</u> <u>Concentration (mg/kg)</u>	<u>Hyperaccumulator Concentration</u> (mg/kg)
Amaranth			
No hyperaccumulator control	249	0.6	
	279	0.8	
	313	1.0	
Average:	281±32.0	0.8±0.2	
Amaranth with Sunflowers	359	1.2	6.8
	339	0.4	5.7
	387	1.0	9.6
	351	0.9	5.2
	336	1.1	13.0
Average:	355±20.4	0.9±0.31	8.1±3.24
Amaranth with Mustard Greens	332	0.2	
	377	1.2	5.0
	351	1.1	
	347	1.4	
Average:	352±18.7	1.0±0.53	5.0±0

Although we initially requested lead contaminated soil at two concentrations, 350 mg/kg and 450 mg/kg, after analyzing the lead content in the soil, it was apparent that the concentration of the ‘high’ lead content soil was much higher than 450 mg/kg. As a result, we have presented the results for the ‘low’ lead content soil and ‘high’ lead content soil for tomatoes. For the other vegetables, we did not have adequate plant growth during this six week period, possibly due to the high concentration of lead in the soil, which was between 1150-1734 mg/kg (Table 2). In addition, the extremely hot weather conditions during this experiment made growing the plants difficult.

Based on the results from this experiment, we can determine that both the mustard greens and sunflowers will preferentially take up lead over the tomato, carrot and amaranth. The concentrations (on a wet weight basis) measured in the mustard greens and the sunflower were consistently higher than the concentrations measured in the garden vegetables for either the low or high lead content soil. Despite this, we did not see any strong trends demonstrating that the presence of the hyperaccumulators reduced the concentration in the garden vegetables over this 6 week period. For a given soil lead concentration and garden vegetable, concentrations in the target vegetable without hyperaccumulators (positive control) were similar to concentrations measured in vegetables grown in pots that also contained hyperaccumulators. One exception may be the tomato grown in the low lead content soil with mustard greens. The concentration (on a wet weight basis) for the control tomato was 0.3 mg/kg, and the concentration in the tomato grown in the pot containing mustard greens was 0.1 mg/kg. However, with only one replicate, we cannot state whether this is a statistically significant difference.

Between the tomato, carrot, and amaranth, we observed the highest lead concentrations in the carrot. The average lead concentration observed in the carrots was between 1.6-1.9 mg/kg, compared with average concentrations of 0.3-0.7 mg/kg (tomatoes grown in low lead content soil) and 0.8-1.0 mg/kg for amaranth. This indicates that root vegetables may be more susceptible to lead contamination compared with shoots or fruits. These concentrations may be compared to the limit of 0.5 mg/kg has been established by the FDA for lead in glazed

dinnerware. This limit is typically applied to lead concentration in food. Based on this limit, the carrot, some samples of tomato and amaranth would be over this limit.

LIMITATIONS

This study was hindered by several obstacles. First, the lead concentration in one batch of soil obtained from the EPA was unexpectedly high compared to the target soil lead concentrations. This not only made comparisons across different groups difficult but also provided difficult growing conditions. Also leading to difficult growing conditions was the high clay content of the soil as well as the above normal temperatures experienced during the growing season. It was decided not to amend the soil to better represent typical growing conditions in Nebraska's backyard gardening community. However, this, coupled with the extreme temperatures provided very difficult growing conditions. This resulted in only a single sample for some of our experiments, and one recommendation for future work would be to repeat this analysis with increased replication, so that statistical significance can be determined for differences observed in lead concentrations in vegetables grown with and without the presence of hyperaccumulators.

IMPLICATIONS

This study indicates that for some vegetables commonly planted in backyard gardens, lead uptake can be reduced by planting lead hyperaccumulating plants. The use of hyperaccumulators is a non-invasive solution to minimizing lead exposure through the ingestion of vegetation grown in contaminated soil. This practice also has the added benefit of minimizing recontamination of raised beds by wind born particles via lead extraction from the soil matrix by the hyperaccumulators. With additional research, using hyperaccumulators may prove to be an easy and inexpensive tool to reducing lead exposure in humans.

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