

Lead Contamination in Saratoga Springs and Hormone-Stimulated Phytoextraction as a Solution

Charles Lovejoy and Úna Semar

Environmental Studies Senior Capstone

Skidmore College, Saratoga Springs, NY

May 10, 2015



Abstract

Lead contaminated soils are a pervasive issue in many urban settings. This study tested lead concentrations in home garden soils of Saratoga Springs residents. Participants were also surveyed about their prior knowledge regarding exposure to lead contamination through produce grown in their home gardens. As a potential solution to this lead contamination, various hormone treatments were used on dwarf variety sunflowers to improve the efficiency of lead phytoextraction. Significant lead contamination was found in over half of the home gardens sampled. The majority of participants had no knowledge of the significant effects of exposure to lead. Sunflowers treated with strigolactone had significantly elevated concentrations of lead in their aboveground and root tissues compared to the other hormone treatments and control group. Future research should be directed toward quantifying the direct human exposure to lead as well as other types of heavy metal contamination. Work should also be done to identify the minimum effective concentration of strigolactone that can be used for phytoextraction in sunflowers.

Introduction

Anthropogenic soil contamination is a major issue facing the modern world. This type of pollution can take the form of heavy metals such as lead and arsenic or other non-biodegradable chemicals like PCB's (USDA 2000). Humans, like all organisms, have always created waste, but our capacity to permanently inundate soils has been facilitated by industrialization and the use of synthetic chemicals. Contamination of terrestrial ecosystems can be caused by a plethora of human activities ranging from manufacturing, mining, and nuclear fission to the use of lead-based paints and persistent organic pesticides (Ross 1994). Soil pollution is so long lasting because unlike air and water contamination, which can disperse easily until they reach non-toxic concentrations, pollutants deposited on land will often remain there until humans are forced to remediate it (Sherene 2010).

Soil contamination has severe implications for public health. Heavy metals like lead, arsenic, copper, mercury, and cadmium are often found in polluted soils. Although these elements are found naturally throughout the world, they can become highly concentrated in certain areas from human industrial activities like mining and engine exhaust (Ross 1994, Sherene 2010). When concentrations of heavy metals reach a particular threshold, they can become hazardous to plants and animals (including humans) that come into contact with the contaminated soils. Extensive exposure to lead, for example, can lead to brain damage, impaired kidney function, and bone degradation (Demayo et al., 1982). Arsenic poisoning, or arsenicosis, can cause diabetes, cardiovascular disease, neurological impairment, and many types of cancers (Kapaj et al., 2007).

Polluted soils are also a major environmental justice issue, as low-income and minority communities are disproportionately exposed to them (Pastor et al., 2002). First brought to public

attention in 1982 when a black community in Warren County, NC was forced to host a toxic waste dump, the environmental justice movement seeks to halt this disproportionate exposure to soil contamination (NRDC 2000). Groups like the NAACP and NRDC have called for a resolution to this issue and continue to urge the government to remediate polluted sites for communities that do not possess the resources to do it themselves.

Soil contamination also threatens the human population's ability to support itself. With the world population currently approaching 7.3 billion and an estimated population of nearly 10 billion by 2050, food production will need to increase by 70% if we are to feed these new mouths (FAO 2011). However, one of the biggest obstacles standing in the way of this is the loss of arable land due to pollution. According to the United Nations Food and Agricultural Organization, 10 million hectares of farmland are lost annually as a result of contamination and overall decline of soil quality (FAO 2011). If the human race is to avoid mass famine, polluted farmland must be remediated and put back into cultivation.

Until recently, there have been two common ways in which soil contamination has been addressed. The first is to cover the polluted soils with a plastic sheet or concrete and wall off the area to prevent human exposure (Alaska DEC 2009). Although inexpensive and quick, this method does nothing to clean up the contamination and has left innumerable abandoned and unusable toxic sites across the globe. The second method, which is exponentially more labor-intensive and expensive, is to dig up the contaminated soils (often amounting to metric tons) and store them in isolated and dry locations such as abandoned mine shafts in Nevada. Clean soil is then imported to replace the removed contaminated substrate (Alaska DEC 2009). In addition to the huge costs of labor and capital, this "solution" to pollution still results in a net loss of soils

that could be used for agriculture and allows for the possibility of spills as the polluted soils are transported to a dumping site.

An alternative method for cleaning up contaminated soils is phytoextraction. This process utilizes specialized plants called hyperaccumulators. These organisms are tolerant to a variety of pollutants and can grow in contaminated soils (Salt et al., 1995). As the plants grow, they absorb the pollutants, like heavy metals or PCB's, and accumulate them in their tissues. Then, at the end of the growing season, the plants are collected, incinerated to decrease the mass of waste, and stored in toxic waste facilities. This process can be repeated over the course of multiple growth seasons, with the end goal of leaving behind uncontaminated and arable soil. This method is less expensive than digging up contaminated soils and does not require the import of clean soils from other locations, thus increasing the amount of arable available for cultivation (Salt et al., 1995).

Scientists have sought to stimulate the phytoremediation process by using phytohormones. Comparable to hormones like testosterone and estrogen in humans, these are chemicals that induce a variety of morphological and chemical changes in the tissues of higher plants and even in the surrounding environment (Tran and Pal 2014). Phytohormones can be distributed into 9 classes: auxins, cytokinins, gibberellins, abscisic acid, ethylene, brassinosteroids, strigolactones, salicylates, and jasmonates (Tran and Pal 2014). In nature, plants produce many different phytohormones at a time. Usually, it is the relative concentration of a hormone in comparison to another hormone that determines the effect. The actions of the hormones used in previous phytoremediation studies and their effect on contaminant absorption can be found in table 1. One of the few hormones not yet studied in phytoremediation experiments is strigolactone. This class of phytohormones stimulates the metabolism and growth

of mycorrhizae, fungi and soil microbes that form symbiotic relationships with plants and their roots (Besserer et al 2006; Besserer et al 2008). Mycorrhizal communities are essential partners of plants and help to absorb water, nutrients and ions from the surrounding soil (Tran and Pal 2014). Because lead is a cation, increasing the growth of mycorrhizal fungi with strigolactones may increase lead extraction rate. Many studies have used these phytohormones in conjunction with chelators such as EDTA, which increase the solubility of lead and make it more bioavailable to phytoextractors (Liphadzi et al 2010; Hadi et al 2014; Tassi et al 2008).

Table 1: actions of certain phytohormones and effect on phytoremediation process		
Phytohormone	Actions in plants	Effect on Phytoremediation
Auxins	Regulates cell growth, and expansion; phototropism and gravitropism; stimulates root growth (Tran and Pal 2014)	Increases extraction efficiency of Mn, Ni, pb and Cd in <i>Helianthus annuus</i> with EDTA chelator (Liphadzi et al., 2010)
Cytokinins	Regulates cell division and differentiation; stimulates shoot bud growth and slows protein catabolism (Tran and Pal 2014)	Increases lead phytoextraction efficiency up to 890% and zinc phytoextraction efficiency by up to 330% in <i>Helianthus annuus</i> (Tassi et al., 2008; Cassina et al., 2012)
Gibberellins	Seed germination, stem elongation, flower development, leaf senescence (Tran and Pal 2014)	GA3 and EDTA-treated <i>Parthenium hysterophorus</i> absorbs up to 8x more cadmium than control (Hadi et al., 2014)
Strigolactones	Stimulate growth of mycorrhizal fungi, induce <i>Striga</i> germination (Besserer et al 2006; Besserer et al 2008)	Not yet studied

One of the plants most commonly utilized in phytoextraction is *Helianthus annuus*, commonly known as the sunflower. This organism can grow in soils contaminated with various heavy metals (Tassi et al., 2008). As fast-growing annuals, sunflower plants can accumulate

hazardous cations in their tissues over the course of the growing season, after which they are removed from the soil, incinerated, and properly disposed of as hazardous waste (Cassina et al., 2012). This process can be repeated over several growing seasons until the soil's heavy metal concentrations reach a safe level.

This study will use a dwarf variety of *Helianthus annuus* to extract lead from contaminated soils treated with EDTA in the Dana Greenhouse at Skidmore College. Plants will be treated with auxin, cytokinin, gibberellin, strigolactone, or a cocktail of all 4 hormones. A control group will also be present. The three goals of this study are to replicate previous phytoextraction work with auxins, cytokinins, and gibberellins; to ascertain if strigolactone can stimulate lead uptake; and also evaluate the efficacy of lead phytoextraction using all four hormones at once.

Methods

Part I: Assessing Lead Contamination in Saratoga Springs

Survey

A sample of twenty-three Saratoga Springs community members who manage a garden at their residence and grow and eat their own produce completed either an online survey or a semi-structured interview. 10 participants completed the online survey through Qualtrics, while 11 participants completed a semi-structured interview. Survey questions (see Appendix I for a list of questions) were administered through semi-structured interviews. Participants who are unavailable for the semi-structured interview will be administered the survey through the Qualtrics online survey platform in digital format. In the case that participants do not have access to the Internet or do not want to complete the survey online, they will either be offered use of the surveyor's laptop computer to take the survey or provided a printed copy that will later be entered online by the surveyor.

Community members were selected through convenient sampling, by sending out emails to individuals with known gardens and taking the first twenty-three responders. The survey examined each participant's current knowledge of soil contamination by heavy metals such as lead (Pb; a common contaminant in urban soils), as well the health effects that can occur from eating produce from soils contaminated with heavy metals, as well as any possible sources of contamination on their property. Community members were also surveyed regarding their current knowledge of remediation techniques for cleaning up soil contamination.

Soil Sampling

Community soil samples from each of the twenty-three household gardens were taken by compositing 7 soil cores systematically taken to a depth of 10 cm using a 4 cm diameter PVC core. Composite samples were then passed through a 2 mm mesh sieve to further homogenize and remove roots, rocks, and large organic material. From each composite sample, three replicate 0.500 g sub samples were dried to a constant mass at 105 °C, the mass recorded, and the dry sample stored in a desiccator with desiccant.

Soil Lead Analysis

Each replicate of the composite sample associated with each household garden was digested using 10 mL of pure nitric acid and a microwave digester (Sarojam, 2011). Digested samples were filtered and then analyzed for Pb concentration on an Atomic Absorption Spectrophotometer in the Skidmore SAIL facility (Sarojam, 2011). Lead concentration was scaled to a per gram of dry soil basis. A one-way ANOVA test was performed to determine if our treatments significantly influenced lead uptake from soil. Results were compared to the EPA standards for acceptable lead content in soil of 300ppm (Environmental Protection Agency, 2015).

Part II: Hormone-Stimulated Phytoextraction of Contaminated Soils

Phytoextraction Set-Up

80 lbs of lead-free, unsterilized topsoil was amended using lead acetate ($\text{Pb}(\text{C}_2\text{H}_3\text{O}_2)_2$) and homogenized (Gleeson 2007). Subsamples were analyzed using atomic absorption spectrometry (Tüzen 2003) in the Skidmore SAIL laboratory. The soil was determined to have an average lead concentration of 2,613 ppm, well above the EPA hazardous threshold of 1,200 ppm (USEPA 2001). This contaminated soil was then transferred into six 10-well planting trays, with a separate planting tray being used for each hormone treatment. Wells contained approximately 420 g of contaminated soil, each of which was then treated with 67.2 mg of EDTA (Tassi 2008; Liphadzi et al 2006) to increase the bioavailability of the lead. Dwarf sunflower seeds (Sunny Smile variety) were sterilized with 70% ethanol and 10% NaClO (Sauer and Burroughs 1986). These were then germinated in peat plugs under grow lights. After 2 weeks of growth, one seedling was transplanted into each well in the planting trays. The sunflowers were then grown in the Skidmore College greenhouse under stable conditions for 4 weeks (see table 2).

Table 2: Growing Conditions in Skidmore Greenhouse

Light Regime	Hours of light/day:12.8 in beginning of April
Temperature	Day: 28-32° C, Night 25-28° C
Humidity	50%
Water	Lightly watered daily
Fertilizer	Neptune's Harvest 2-4-1 NPK Fertilizer; initial fertilization = 1oz fertilizer/1gallon water; subsequent fertilization (2 weeks later) = 0.5oz fertilizer/1gallon water
Pest control	Manually remove visible pests upon daily watering

Application of Phytohormone Solutions

IAA (auxin), Kinetin (cytokinin), and GA (gibberellin) were obtained from sigma Aldrich. GR24 (strigolactone) was obtained from StrigoLab at Turin University (Italy). Phytohormone treatments (see table 2) were applied to the sunflowers growing in the experimental treatment groups on the 7th, 9th, and 11th days after being transplanted into the lead-contaminated soils. The IAA (auxin), GA3 (gibberellin), and Kinetin (cytokinin) solutions were applied using foliar sprays, while the GR24 (strigolactone) was added by pouring 15mL of solution at the base of each plant (Tassi et al 2008; Hadi et al 2014; Liphadzi et al 2006; Besserer et al 2008).

Table 3: Phytohormone Treatment Groups

	Chemical	Concentration	Application Method	Volume Applied
Control	DI water	n/a	Foliar spray	Wet all visible tissue
Auxin	Indole acetic acid (IAA)	25 mg/L	Foliar spray	Wet all visible tissue
Cytokinin	Kinetin (6-Furfuryl-aminopurine)	20 mg/L	Foliar spray	Wet all visible tissue
Gibberellin	Gibberellic acid (GA3)	35 mg/L	Foliar spray	Wet all visible tissue
Strigolactone	GR24	0.02685 mg/L	Application to soil at plant base	15 mL
All Hormones	All four hormone treatments above will be performed at the same time to every plant in this treatment group			

Harvesting and Lead Analysis

Two weeks after the last hormone treatment, 6 plants were randomly selected from each treatment group and harvested. The aboveground tissues were cut using shears, while the root systems were separated from the soil and washed. Both aboveground and root tissues were dried

at 80 °C. The tissues of each sunflower were weighted and then analyzed for lead content using AAS after digestion by nitric acid in a microwave digester (Sarojam, 2011). Lead concentration was scaled to a ug Pb per gram of dry tissue basis and then an average lead content was calculated for the roots and aboveground tissue in each treatment group. A one-way ANOVA test was performed in JMP to determine if our treatments significantly influenced lead uptake from soil.

Results

Part I: Assessing Lead Contamination in Saratoga Springs

Survey and semi structured interview data

A total of 23 Saratoga Springs residents participated in this study, however, only 21 out of these 23 people fully completed either an online survey or a semi-structured interview. 10 participants completed an online survey, while 11 participants completed a semi-structured interview.

Community prior involvement in home garden soil testing

The public was largely uninvolved in any type of soil testing prior to this study. 86% (18/21) of participants had never tested their garden soil at all. 14% (3/21) of participants had tested their garden soil prior to establishing home gardens (Figure 1). However, this testing was primarily for pH levels. When asked about their involvement in soil testing for lead concentrations specifically, respondents largely indicated that they had never engaged in lead testing of their garden soil. Specifically, 95% (20/21) of participants had never tested their garden soil for lead concentrations, while 5% (1/21) of participants had tested their garden soil for lead concentration prior to establishing a home garden (Figure 2).

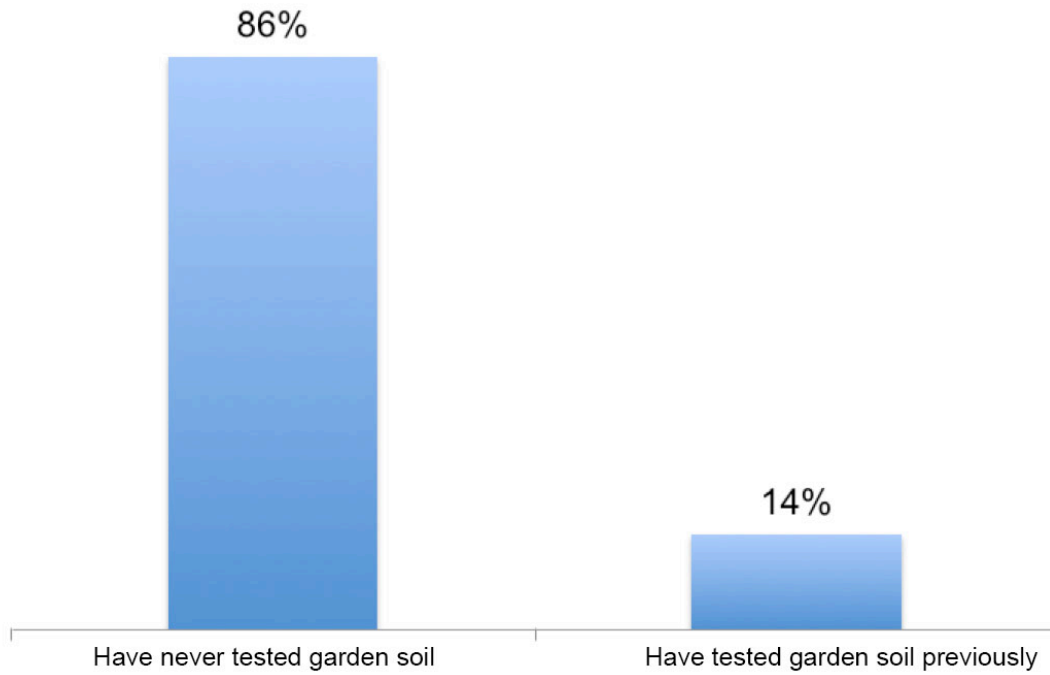


Figure 1. Percentages of participants who have engaged in any type of garden soil testing

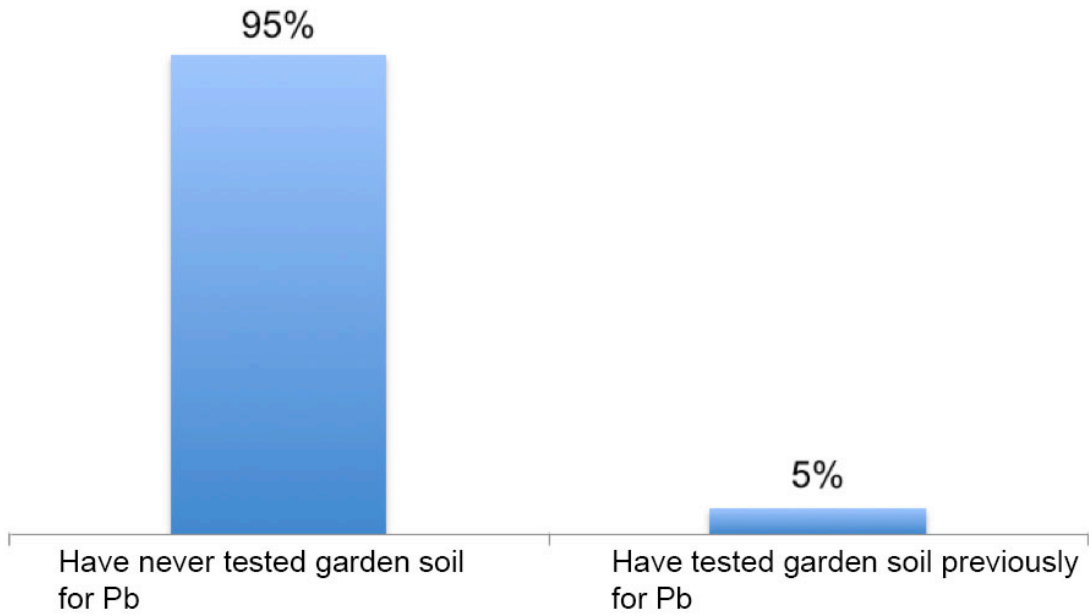


Figure 2. Percentage of participants who have tested garden soil for lead concentration

Community Awareness

Regulatory standards for lead contamination

95% (20/21) of respondents indicated that they were unaware of the EPA regulatory standards regarding safe levels of lead concentrations in home garden soil that is being used to grow produce. 5% (1/21) of participants were aware of these regulatory standards (Figure 3).

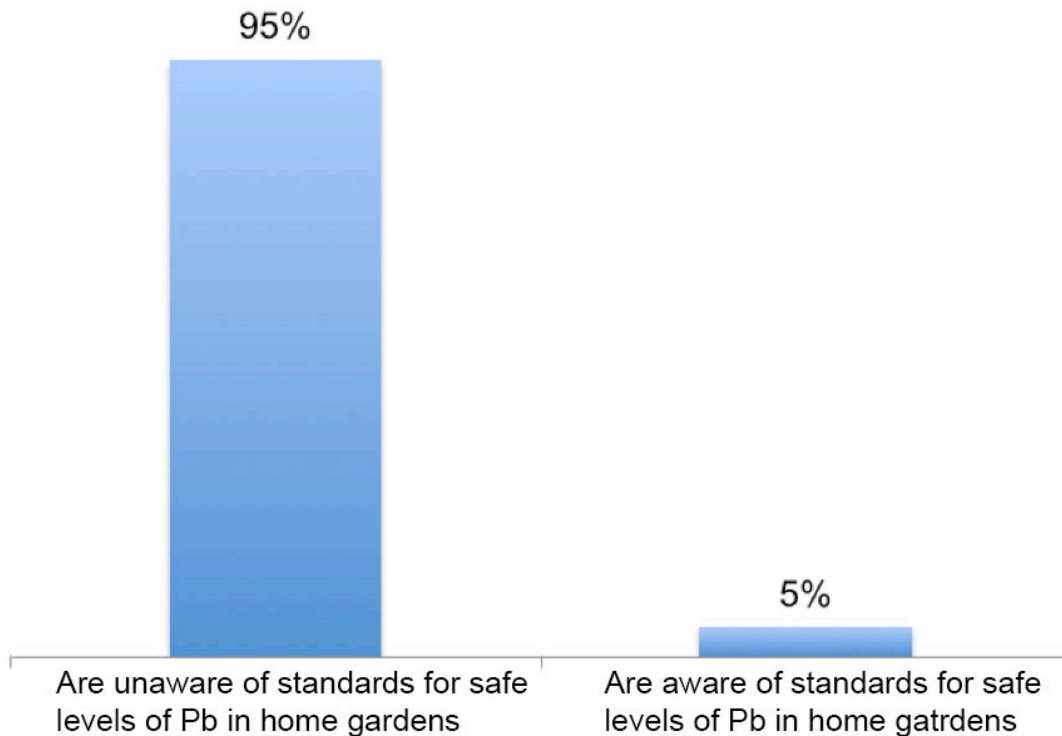


Figure 3. Participant’s awareness of standards for safe levels of lead in home gardens

Health risks

67% (14/21) of participants were unaware of the health risks associated with exposure to lead contamination. Comparatively, 33% (7/21) participants were aware of the health risks associated with exposure to lead contamination. One respondent explained “I know that children are most vulnerable to lead exposure, but I do not know specific health effects” (personal communication, 2016). Similarly, another respondent explained “I know nothing about it”

(personal communication, 2016). Further respondent representative quotations regarding awareness of health risks associated with lead exposure are presented below in Table 4.

Possible sources of lead contamination

48% (10/21) of participants were unaware of possible sources of lead contamination on their own property. 52% (11/21) of participants were aware of possible sources of lead contamination on their own property, and cited lead based paint as a primary concern. One respondent explained “Our house was built in the 1890s, so we are sure that lead paint has been an issue on our property” (personal communication, 2016). Similarly, another respondent explained “Lead paint...our house predates 1978. We have replaced all the old windows, but saved some...we used a few to make a cold frame but dismantled it after a year” (Qualtrics, 2016). Further respondent representative quotations regarding awareness of possible sources of lead contamination are presented below in Table 4.

Remediation techniques

86% (18/21) of respondents were unaware of remediation techniques for cleaning up lead contaminated soil. Comparatively, 14% (3/21) of participants were aware of only conventional methods of cleaning up lead contaminated soil by excavation. 100% (21/21) of participants were unaware of newer methods of remediation of lead contamination, such as phytoextraction. Respondent representative quotations regarding awareness of remediation techniques for cleaning up lead contamination are presented below in Table 4.

Health Risks	Sources of lead contamination	Remediation techniques
I don't know very much about the health effects associated with lead exposure	There was an old house on the site and they dumped a lot of car parts, etc. This is why we use raised beds!	At this time I do not know much about lead removal from soil
I have no knowledge about lead side effects	Our house was built in the 1890s, so we are sure that lead paint has been an issue on our property	No knowledge of this on my end
I know nothing about it	The house was built in a wooded area after lead was banned from paint. Some soil was brought in to help level the backyard. The source of that soil is not known	I don't know any way to clean up lead contamination, but if my soil is found to be contaminated I would do extensive research on the internet to educate myself
I know that children are most vulnerable to lead exposure, but I do not know specific health effects	Lead paint...our house predates 1978. We have replaced all the old windows, but saved some...we used a few to make a cold frame but dismantled it after a year.	I don't know specifically how to remediate contaminated soil.
I know that lead is a neurotoxin that if consumed by children it affects brain functions, such as learning and behavior.	Lead paint was likely used on our house before it was banned, dust from lead paint is a problem for everyone	No knowledge about how to clean up contaminated soils, but constructing raised beds and importing soil may be an option

Table 4. Representative quotations from respondents

Soil lead concentrations

61% (14/23) of the soil samples taken from home gardens were severely contaminated with lead, with concentrations ranging from 104 ppm to 869ppm. 39% (9/23) of the soil samples had lead contamination present at lower levels, ranging from 29 ppm to 87 ppm (Figure 4).

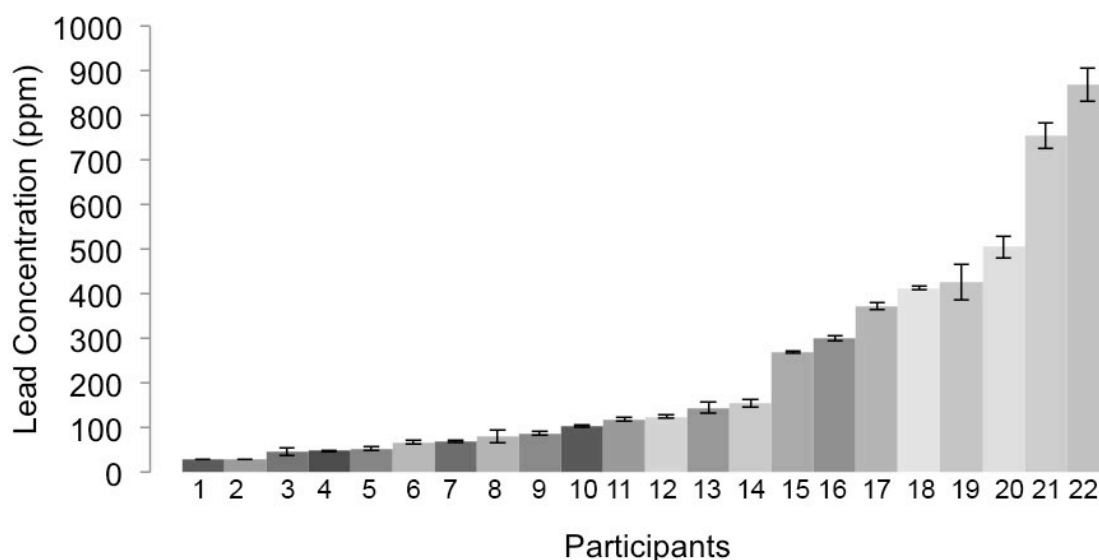


Figure 4. Lead concentrations ranging from 0-1000 ppm for individual home gardens in Saratoga Springs, NY

Part II: Hormone-Stimulated Phytoextraction of Contaminated Soils

A one-way ANOVA test was run using the results from the aboveground tissue analysis to determine if the average lead concentrations in each treatment group were statistically different from one another. The average lead concentration for the control, auxin, and gibberellin treatments (group A) were not significantly different from each other (1,428 +/- 68.7 vs 1,439 +/- 77.2 vs 1,127 +/- 230.4; M +/- SE). The average lead concentration for the cytokinin treatment (2,253 +/- 198.5; M +/- SE) (group B) was significantly higher than all treatments in group A. The average lead concentration for the strigolactone and all hormones treatments (4,603 +/-

101.7 vs 4,916 +/- 130; M +/- SE) (group C) was significantly higher than treatments in groups A and B. $F=125.27$, $P<0.0001$. A Tukey-Kramer HS Comparison of Means was run to conclude that the pattern of variance was driven most strongly by strigolactone.

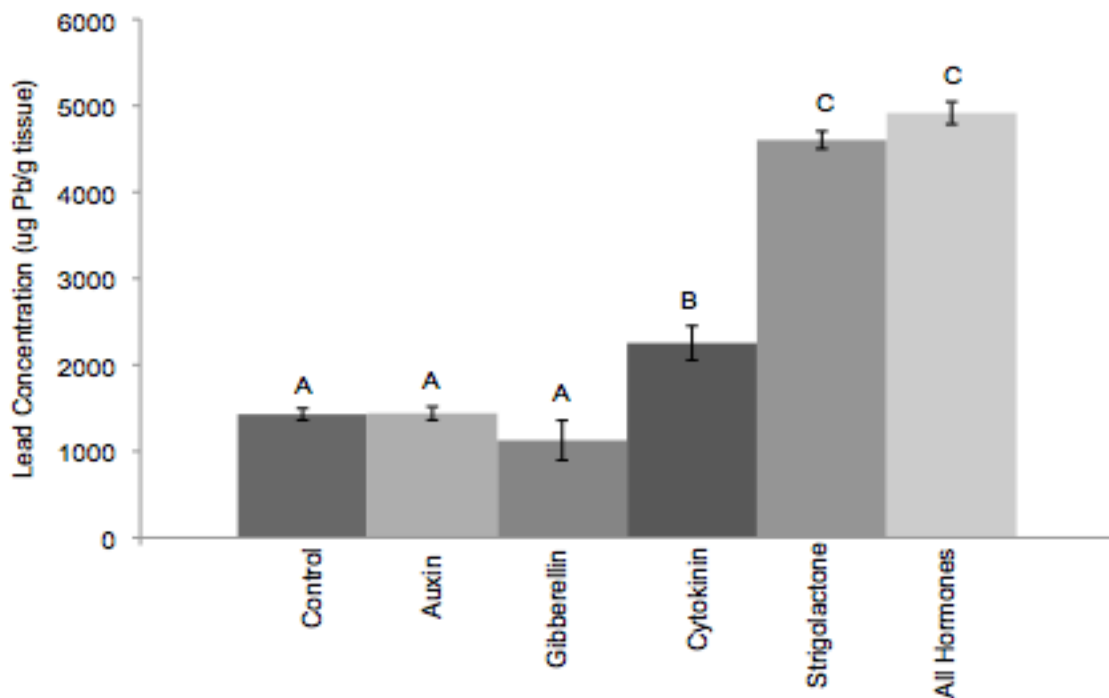


Figure 4: Average Lead Concentrations in Aboveground Tissues

The strigolactone and all hormones treatments had significantly higher lead concentrations compared to the cytokinin treatment. The cytokinin treatment had significantly higher lead concentrations compared to the control, auxin, and gibberellin treatments.

Statistical analysis was not performed for the lead concentrations in the root tissues, as these were analyzed as composite samples. However, the same general pattern was observed in

the roots as in the aboveground tissues, with the cytokinin, strigolactone, and all hormones roots having higher lead concentrations than the control, auxin, and gibberellin groups.

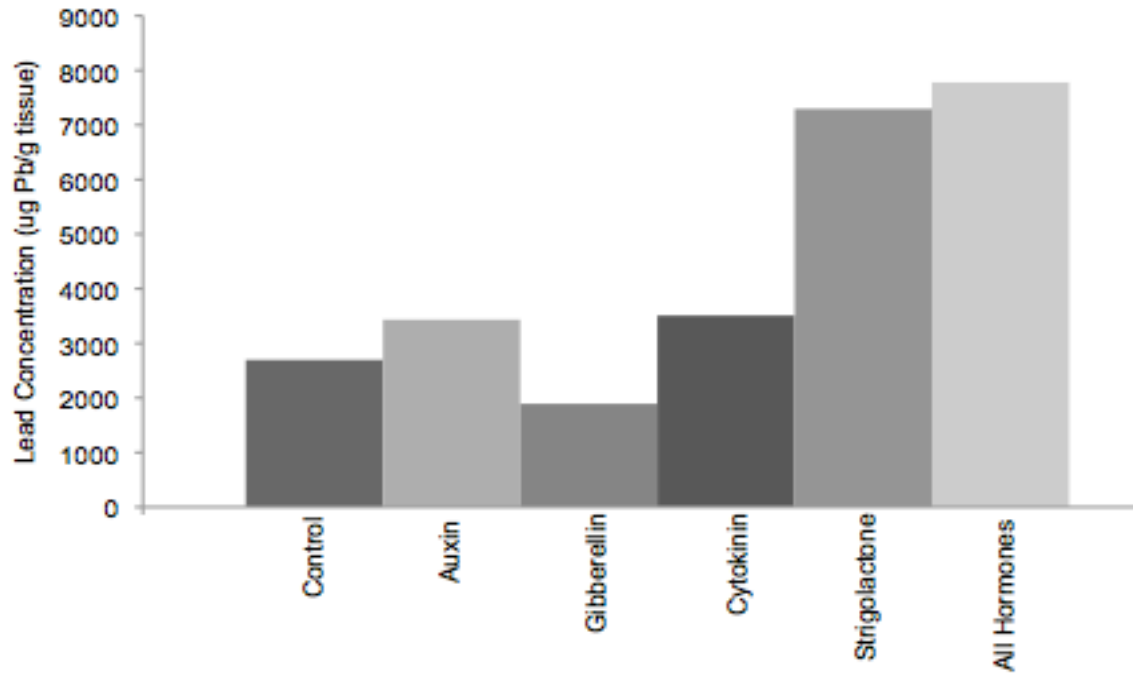


Figure 5: Average Lead Concentrations in Root Tissues

The strigolactone and all hormones treatments had higher lead concentrations compared to the control, cytokinin, auxin, and gibberellin treatments. Overall lead concentrations are higher in the roots than in the aboveground tissues.

Significant morphological changes were also observed with belowground tissues, with strigolactone plants exhibiting significantly larger root systems than the plants treated with the other hormones



Figure 6: Morphological Changes in Root Biomass between Strigolactone and Gibberellin Treatment Groups

Plants treated with strigolactone had significantly larger root systems than plants treated with gibberellin. This difference was apparent, although not as significant, between strigolactone and the other hormones.

Discussion

Part I: Assessing Lead Contamination in Saratoga Springs

Community awareness and knowledge regarding potential exposure to lead through contaminated garden soil was found to be lacking tremendously. Most importantly, members of the public who participated in this study were generally unaware of the importance of testing their soil for contamination prior to growing and consuming potentially contaminated produce. Participants were also generally unaware of the health risks posed by potential exposure to lead, as well as ways to clean up lead contaminated soil. This general lack of awareness amongst participants indicates a concerning trend that is not isolated in nature. Grossman (2016) suggests that enthusiastic gardeners in other urban areas, such as Portland, often do not ask questions

about the safety of their backyard soil prior to gardening. Often, the appeal of establishing a home garden, including the ability to decrease the amount of money spent on groceries, the ability to engage in healthy and sustainable living and to engage with nature, often precedes the appeal of spending money on testing home soils. Grossman (2016) also suggests that in many semi-urban areas, where industrial legacies are less observable, residents tend to blindly trust the safety of their soils.

Community soil testing revealed that the majority of the home gardens sampled in this study were severely contaminated with lead, demonstrating that lead contamination is a pervasive issue within the Saratoga Springs community. This is significant, as Saratoga Springs is a relatively affluent semi-urban area, and lead contamination is most frequently associated with lower income urban areas (Pastor et al., 2002, Evans & Kantrowitz, 2002).

The results of this study suggest that education and awareness regarding lead contamination in home gardens, even in areas in which this type of contamination is an issue, is severely lacking. In order to make broader conclusions regarding the pervasiveness of this issue, further studies should include a larger, more representative sample size for home gardens within Saratoga Springs. In order to determine the actual extent of exposure of participants, it may also be interesting for future research to test produce grown in contaminated gardens to see if lead is being transferred from these soils to people.

Part II: Hormone-Stimulated Phytoextraction of Contaminated Soils

Statistically significant differences in lead accumulation by sunflowers were caused by the diverse hormone treatments. Most importantly, strigolactone, which has not been extensively studied in phytoextraction experiments, significantly increased lead concentrations in the sunflowers. The strigolactone also appeared to increase root biomass. Both of these responses

might be incited by a stimulation of the mycorrhizal communities, which has been suggested by previous studies as a primary action of strigolactone hormones (Besserer et al 2006; Besserer et al 2008). It is important to note that because the soils were contaminated using $\text{Pb}(\text{C}_2\text{H}_3\text{O}_2)_2$, there was a significant amount of acetate in the soil. Microbes, fungi, and plants can readily use this simple sugar. Thus, additional stimulation of the mycorrhizae may have occurred as a result of the lead (II) acetate. However, the $\text{Pb}(\text{C}_2\text{H}_3\text{O}_2)_2$ was equally spread throughout the treatment groups due to the homogenization, so while the overall lead accumulation in this study may have been higher than it would have in the absence of the acetate addition, the individual differences between hormone treatments cannot be discounted.

The average lead concentration in the all hormones treatment was higher than, but not statistically different from, the average for the strigolactone treatment. This could be caused by a synergy between the cytokinin and strigolactone, both of which incited greater lead accumulation compared to the control.

Auxin, cytokinin, and gibberellin have all been shown by previous studies to significantly increase lead accumulation compared to controls in a variety of hyperaccumulating plants, including *Helianthus annuus* (Liphadzi et al., 2010; Tassi et al., 2008; Cassina et al., 2012; Hadi et al., 2014). Our study only replicated this result for cytokinin, though this was still by a smaller margin than anticipated. This may be due to the fact that the sunflowers in this study were only allowed to grow in the contaminated soils for a total of 4 weeks due to time constraints, whereas this dwarf variety has a full growth cycle that is over twice as long. Had we allowed the sunflowers to grow for another month, enhanced differences in lead accumulation may have been observed between the auxin, cytokinin, and gibberellin treatments. It is also possible that these three hormones do not have the same potency in dwarf organisms as they do in regular varieties.

The results of this study, particularly the enhanced accumulation of lead caused by the strigolactone treatment, suggest that the efficiency of the phytoextraction process can continue to be improved. Further studies must be performed to verify these results. It will also be prudent to experiment with different concentrations of GR24 (as well as other strigolactone compounds) to determine the lowest concentrations that can be used while still maintaining its stimulating effects. GR24 is currently expensive due to it being recently discovered and only synthesized by a handful of specialty labs. Thus, at this time, it is not cost-effective to be used on a large scale, although this may change as production of GR24 and other strigolactone compounds expands. It is our hope that with continued work, phytoextraction will become a viable alternative to excavation in the cleanup of contaminated soils.

Appendix I. Survey Questions

1. Have you taken any courses, workshops or attended any public talks about agriculture, the environment or conservation? If so, can you specify what kinds of courses?
2. Do you read books/newspapers or watch movies about agriculture, the environment or conservation? If so, can you name them?
3. What experiences do you have with gardening/food production? What herbs/fruits/vegetables do you grow?
4. What percentage of home needs come from your garden?
5. Have you encountered any problems with your soil quality (e.g., wilting, rust on cucumbers, fertility)? If so, what?
6. Have you ever tested any of the soils on your property? If so, for what?
7. Are you aware of any possible sources of lead (Pb) contamination on your property? If so, what are they?
8. What do you know about lead contamination and its health effects?
9. Are you aware of any standards/regulations regarding “safe” levels of lead in home gardens?
10. If your soil is found to be contaminated, what kinds of steps would you take?
11. Can you discuss any knowledge you have regarding cleaning up soil contamination?
12. Can you discuss any knowledge you have about phytoremediation?
13. Do you think that agricultural production within city limits poses significant human health risks?

References

- Alaska Department of Environmental Conservation. 2009. Environmental cleanup methods.
- Alonso, E., Cambra, K., Martinez, T. 2001. Lead and cadmium exposure from contaminated soil among residents of a farm area near an industrial site. *Archives of Environmental Health*. 56;3. 278.
- Audet P. 2011. Arbuscular mycorrhizal symbiosis and other soil interactions in relation to environmental stress. *Environmental Adaptations and Stress Tolerance of Plants in the Era of Climate Change*: 233-264.
- Brown, K.H., Jameton, A.L. 2000. Public health implications of urban agriculture. *Journal of Public Health Policy*. 21;1. 20-39.
- Besserer A, Puech-Pagés V, Kiefer P, Gomez-Roldan V, Janeau A, Roy S, Portais JC, Roux C, Bécard G, Séjalon-Delmas N. 2006. Strigolactones stimulate arbuscular mycorrhizal fungi by activating mitochondria. *PLOS Biology*, 4(7).
- Besserer A, Bécard G, Jauneau A, Roux C, Séjalon-Delmas N. 2008. GR24, a synthetic analog of Strigolactones, stimulates the mitosis and growth of the arbuscular mycorrhizal fungus *Gigaspora rosea* by boosting its energy metabolism. *Plant Physiology*, 148(1): 402-413.
- Cassina L, Tassi E, Pedron F, Petruzzelli G, Ambrosini P, Barbafieri M. 2012. Using a plant hormone and a thioligand to improve phytoremediation of Hg-contaminated soil from a petrochemical plant. *Journal of Hazardous Materials*, 231: 36-42.
- Creswell, J. W., Plano Clark, V. L., Gutmann, M. L., & Hanson, W. E. (2003). Advanced mixed methods research designs. *Handbook of Mixed Methods in Social and Behavioral Research*, 209-240.
- Demayo A, Taylor M, Taylor K, Hodson P, Hammond P. 1982. Toxic effects of lead and lead compounds on human health, aquatic life, wildlife plants, and livestock. *Critical Reviews in Environmental Control* 12(4): 257-305.
- Environmental Protection Agency. 2015. Hazard standards for lead in paint, dust, and soil. United States Environmental Protection Agency.
- Evans, G.W., Kantrowitz, E. 2002. Socioeconomic Status and Health: The Potential Role of Environmental Risk Exposure. *Annual review of public health*. Vol. 23: 303-331
- Farrell, K.P., Merrill C., Brophy, M.S., Chisolm, J., Rohde, PhD, and Strauss, W.J. 1998. Soil lead abatement and children's blood lead levels in an urban setting. *American Journal of Public Health*. 88;12.
- Finster, M.E., Gray, K.A., Binns, H.J. 2004. Lead levels of edibles grown in contaminated residential soils: a field survey. *Science of The Total Environment*. 320;2-3, 245-257.

- Food and Agriculture Organization of the United Nations. 2011. The state of the world's land and water resources for food and agriculture: managing systems at risk.
- Gleeson, A.M. 2007. Phytoextraction of Lead from Contaminated Soil by *Panicum virgatum* L. (Switchgrass) and Associated Growth Responses. Master's Thesis, Queen's University, Kingston, Ontario, Canada.
- Grossman, E. 2016. What Portland's Soil Crisis Can Teach us About Heavy Metals in the Garden. *Civil Eats*. <http://civileats.com/2016/03/08/what-portlands-soil-crisis-can-teach-us-about-heavy-metals-in-the-garden/>
- Hadi F, Ali N, Ahmad A. 2014. Enhanced phytoremediation of cadmium contaminated soil by *Parthenium hysterophorus* plant: effect of gibberellic acid (GA3) and synthetic chelator, alone and in combinations. *Bioremediation journal* 18(1): 46-55.
- Hough, R.L., Breward, N., Young S.D., Crout, N.M.J., Tye, A.M., Moir, A.M., Thornton, L. 2004. Assessing Potential Risk of Heavy Metal Exposure from Consumption of Home-Produced Vegetables by Urban populations. *National Institute of Environmental Health Sciences*. 112;2. 215-221.
- Jacob, D.E., Clickner, R.P., Zhou, J.Y., Viet, S.M., Marker, D.A., Rogers, J.W., Zeldin, D.C., Broene, P., Friedman, W. 2002. The Prevalence of Lead-based paint hazards in U.S. Housing. *Environmental Health Perspectives*. 110;10.
- Kapaj S, Peterson H, Liber K, Bhattacharya P. 2007. Human health effects from chronic arsenic poisoning- a review. *Journal of Environmental Science and Health* 41(10): 2399-2428.
- Lanphear et al. 1998. The contribution of lead-contaminated house dust and residential soil to children's blood lead levels: A pooled analysis of 12 epidemiologic studies. 79;1. 51-68.
- The Lead Group. 1997. Is your yard lead safe? *Lead Action News*. 5;3.
- Liphadzi M, Kirkham M, Paulsen G. 2010. Auxin-enhanced root growth for phytoremediation of sewage-sludge amended soil. *Environmental Technology*, 27(6): 695-704.
- Markus, J., McBratney, A.B. 2001. A review of the contamination of soil with lead: II. Spatial distribution and risk assessment of soil lead. *Environmental International*. 25;5. 399-411
- Mielke, H.W., Reagan, P.L. 1998. Soil is an important pathway of human lead exposure. *Environmental Health Perspectives*. 106. 217-229
- Niinae M, Nishigaki K, Aoki K. 2008. Removal of lead from contaminated soils with chelating agents. *Materials Transactions* 49(10): 2377-2382

- NRDC. 2000. The environmental justice movement. Accessed Nov. 13 2015
<<http://www.nrdc.org/ej/history/hej2.asp>>
- Pastor M, Sadd J, Hipp J. 2002. Which came first? Toxic facilities, minority move-in, and environmental justice. *The Journal of Urban Affairs* 23(1): 1-21.
- Ross S. 1994. Sources and forms of potentially toxic metals in soil-plant systems.
<<http://www.wiley.com/WileyCDA/WileyTitle/productCd-0471942790.html>>
- Salt D, Blaylock M, Kumar N, Dushenkov V, Ensley B, Chet I, Raskin I. 1995.
Phytoremediation: a novel strategy for the removal of toxic metals from the environment using plants. *Nature Biotechnology* 13: 468-474.
- Sarojam, P. 2011. Analysis of Pb, Cd and As in Spice Mixtures using Graphite Furnace Atomic Absorption Spectrophotometry. *Perkin Elmer, Inc.*
- Sauer D, Burroughs R. 1968. Disinfection of seed surfaces with sodium hypochlorite. *Phytopathology* 76(7): 745-749
- Sherene T. 2010. Mobility and transport of heavy metals in polluted soil environments. *Biological Forum* 2(2): 112-121.
- Sipter, E., Rózsa, E., Gruiz, K., Tatrai, E., Morvai, V. 2008. Site-specific risk assessment in contaminated vegetable gardens. *Chemosphere*. 71;7. 1301-1307.
- Tassi E, Pouget B, Petruzzelli G, Barbaferi M. 2008. The effects of exogeneous plant growth regulators in the phytoextraction of heavy metals. *Chemosphere*, 71(1): 66-73.
- Tran L and Pal S. 2014. Phytohormones: a window to metabolism, signaling, and biotechnological applications. Springer Science and Business Media New York.
- Tüzen M. 2003. Determination of heavy metals in soil, mushroom and plant samples by atomic absorption spectrometry. *Microchemical Journal* 74(3): 289-297
- USDA NRCS. 2000. Heavy metal soil contamination. Accessed online Dec. 8 2015
<http://www.nrcs.usda.gov/Internet/FSE_DOCUMENTS/nrcs142p2_053279.pdf>
- US Environmental Protection Agency. 2001. Hazard Standards for Lead in Paint, Dust, and Soil (TSCA Section 403). Accessed online Dec. 8 2015 <<http://www.epa.gov/lead/hazard-standards-lead-paint-dust-and-soil-tsca-section-403>>