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

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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 0 00o 0 nend(c) 0.2 (e) 0.2 d0 00o 0 nire(of) -0.5 (t) 0.2 (e) 0.2 (n f) -0.5 onun(i) 0.2 n(pol) 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,

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 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.

Community members were selected through convenient sampling, by sending out emails to individuals with known gardens and taking the first twenty

80 lbs of lead-free, unsterilized topsoil was amended using lead acetate ($Pb(C_2H_3O_2)_2$) 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).

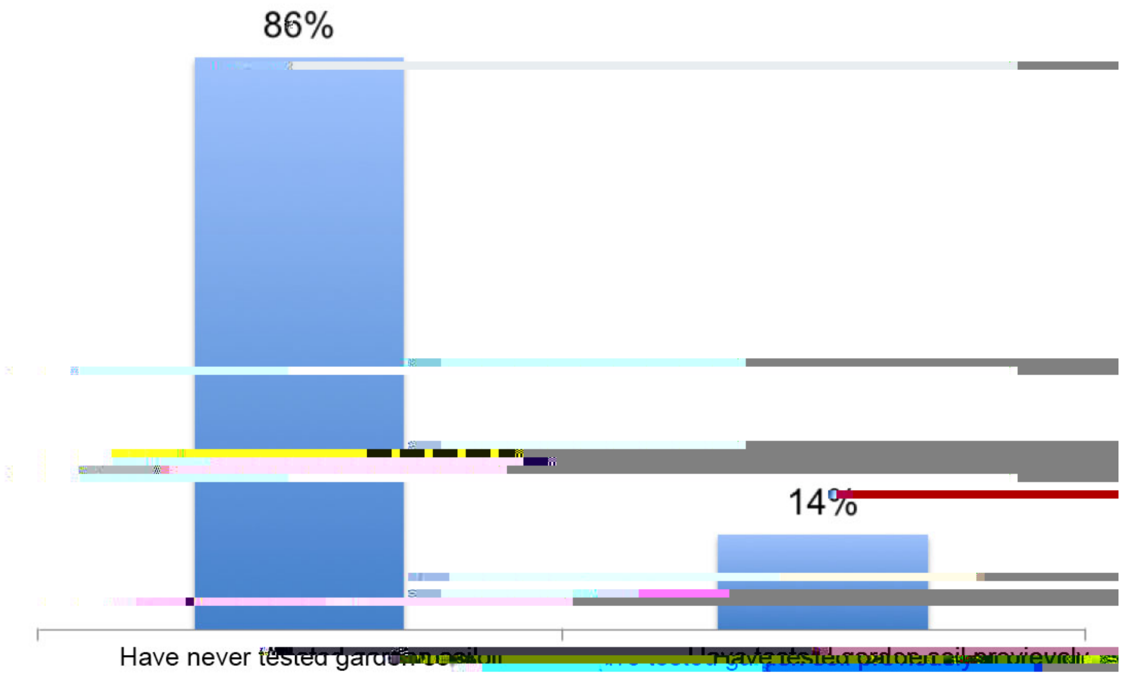
: Growing Conditions in Skidmore Greenhouse

Light Regime	Hours of light/day:12.8 in beginning of April
Temperature	Day: 28-32° C, Night 25-28° C
Humidity	50%
Water	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

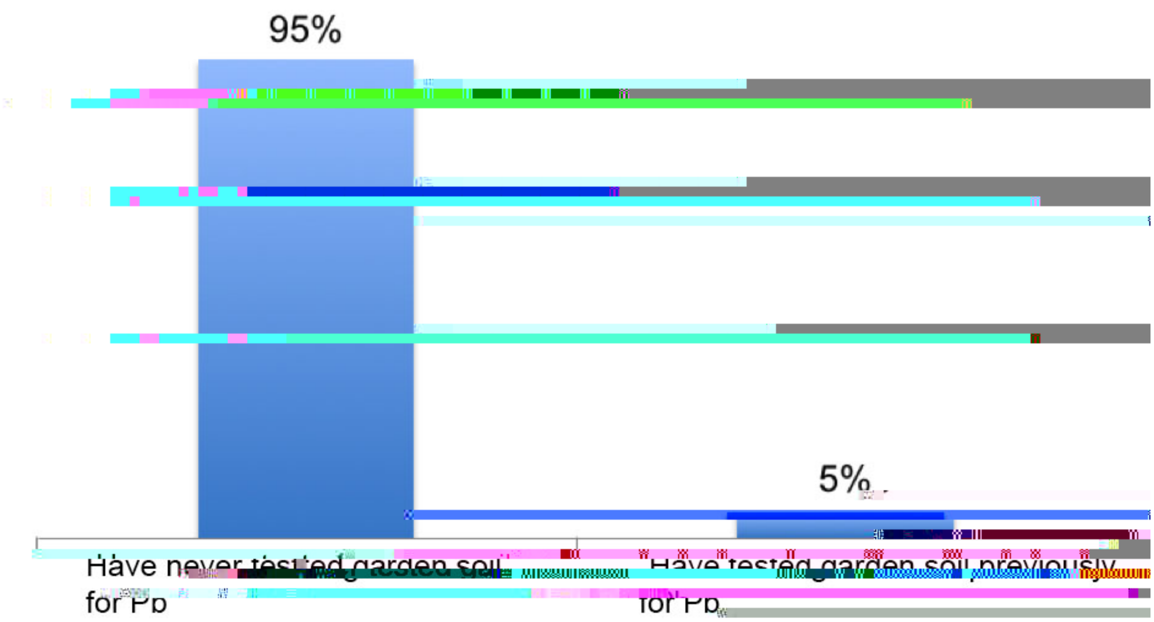
IAA (auxin), Kinetin (cytokinin), and GA (gibberellin) were obtained from sigma Aldrich. GR24 (strigolactone) was obtained from StrigoLab at Turin University (Italy).

Phytohormone treatments (see table 2)

at 8



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



Percentage of participants who have tested garden soil for lead concentration

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).

Participant's awareness of standards for safe levels of lead in home gardens

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"

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	

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).

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

A one-way ANOVA test was run using the results from the aboveground tissue analysis

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.

: Average Lead Concentrations in Aboveground Tissues

The strigolactone and all hormones treatments had significantly higher lead concentrations 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.

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.

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 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 $Pb(C_2H_3O_2)_2$, 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 $Pb(C_2H_3O_2)_2$ 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

The results of this study, particularly the enhanced accumulation of lead caused by the

Survey Questions

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