THE VALUE OF JAPANESE KNOTWEED IN PHYTOREMEDIATION OF CONTAMINATED SOILS ALONG THE WOONASQUATUCKET RIVER

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Thesis

Submitted in partial fulfillment of the
requirements for degree of Master of Arts at the
Center for Environmental Studies,
Brown University, Providence, RI
May 2002

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Abstract

Japanese knotweed (*Polygonum cuspidatum*) was evaluated for its worth as a hyperaccumulator of heavy metals in soils along the Woonasquatucket River, a historically polluted river. Plant tissue, soil and sediment samples were collected from 7 sites along the lower portion of the Woonasquatucket River and analyzed for elemental composition. Lead, copper, chromium, zinc, nickel, and cadmium were analyzed using Atomic Absorption Spectrometry (AA) and Inductively Coupled Plasma Spectrometry (ICP). Results show that Japanese knotweed is tolerant of zinc, lead and copper in soil. However, metal concentration in below and aboveground tissue of knotweed indicate that it does not hyperaccumulate heavy metals from the soil. Further calculations show that knotweed is not effective at removing metals from contaminated soils in a reasonable timeframe. Thus, Japanese knotweed should not be used to remediate contaminated soils along the Woonasquatucket River.

1. **Introduction**

Japanese knotweed (*Polygonum cuspidatum*) is an aggressive perennial plant that is native to northeast Asia. Introduced to the United States in the late 19th century as an ornamental plant, Japanese Knotweed (JK) has spread rapidly throughout the Northeast. Considered a noxious weed in many states, JK inhabits disturbed, derelict and unmanaged land. In Rhode Island, knotweed is represented everywhere from urban parks to roadside margins.

Japanese knotweed is particularly invasive to riparian habitats, and is ubiquitous along the Woonasquatucket River, a historically polluted river. The weed represents a significant component of the plant assemblage in the lower portion of the watershed, where it has out-competed native plant species along the riparian buffer. Restoration efforts by the Rhode Island Department of Environmental Management (RIDEM) and the Woonasquatucket River Watershed Council (WRWC) have focused on restoring the riparian habitat along the Woonasquatucket River. Included in their plan to restore the riparian buffer is planting of native plant species and eradication of Japanese Knotweed.[i]

However, is it wise to remove Japanese Knotweed before knowing how it functions in the ecosystem? Not all invasives are considered useless, i.e. provide some benefits. Japanese Knotweed has been valued for its medicinal attributes as well as for a source of food. It has also been used for erosion control, and is sometimes grown as an ornamental. Thus, rather than expending energy and resources on eradicating Japanese Knotweed, this study looks at ways to maximize resources spent on managing the plant's potential benefits. The benefit that is of most concern to this study is the value of knotweed in phytoremediation. Phytoremediation is an emerging clean-up technology based on the well-known ability of plants to take up and concentrate contaminants in their tissues.

Hence, this study focuses on evaluating the potential of Japanese Knotweed as an effective phytoremediator of heavy metal contaminated soils along the Woonasquatucket River. It does so by addressing the following main research questions: (1) what metals are accumulated in Japanese knotweed, and in what plant part? (2) What is the relationship between soil and plant part concentration? (3) How long will it take knotweed to reduce metal concentration in soil to an acceptable level?

To achieve these research objectives, this paper is divided into 5 sections: The first section outlines background information. Next, the methods used to collect and analyze samples are discussed. The third section focuses on the results of the analyses, while the last section summarizes the major conclusions and outlines recommendations for knotweed management.

2. **Background**

2.1 ***Japanese Knotweed***

2.1.1 *Origin*

Native to Northern Asia, Japanese knotweed (*Fallopia japonica* (Houtt.) Ronse Decraene) is also known by the scientific names of *Reynoutria japonica* Houtt and *Polygonum cuspidatum* Sieb.& Zucc.[ii] Due to the history of its discovery, the taxonomy and nomenclature of this species has been through many changes. Although it is commonly referred to by these three genera, knotweed is now classified under the genus Fallopia (Ronse Decraene and Akerroyd).[iii] A member of the buckwheat family (Polygonaceae), Japanese knotweed was brought over to Europe and North America as an ornamental in the early 19th century.[iv] By turn of the century, Japanese knotweed (knotweed) had become a firmly established ornamental garden plant in the United States. Sold by many nurseries, knotweed was recommended for planting in wet soil by the sides of pond and streams.[v] Shortly after its introduction to the horticulture world, the hardy plant escaped into the wild where its spread throughout the Northeast has been exponential.

2.1.2 *Reproduction and Growth*

Knotweed forms dense thickets, and has an extensive woody rhizome system.[vi] It grows rapidly, reaching up to 10ft at maturity, at a growth rate of 4.65 cm per day.[vii] Knotweed spreads horizontally after establishment forming dense monoclonal stands called patches with diameters of up to 10m.[viii] Knotweed is a deicious perennial plant that reproduces by means of sexual and vegetative growth.[ix] However, in the United States and United Kingdom, vegetative growth is the primary means of reproduction. This is largely because fertile male plants are rarely found in either country. In fact, all knotweed plants found in Europe and North America are believed to be female and DNA studies have shown that they probably originate from the same "mother plant."[x] Even where seedlings have been found in New Jersey, USA, survival there was neglible.[xi] Thus, knotweed plants in the US primarily rely on rhizome fragmentation and budding for propagation. In addition to sending out stems from rhizome, knotweed can also regenerate from stem tissue. A small piece of stem less than 7g can give rise to new plants,[xii] making it difficult to control through mechanical means.

2.1.3 *Distribution and Habitat*

Japanese knotweed's natural habitat in Japan is recent volcanic lava, which it invades within 20 years of activity.[xiii] In the US and UK, it is most commonly found in open habitats such as riverbanks, river islands, disturbed wetlands, roadside margins and in areas with disturbed soils. It is a primary colonizer of unmanaged, derelict land and has been particularly effective at colonizing the banks of urban rivers.[xiv] However, in areas that have not been affected by industry or urbanization, knotweed is absent and is generally less common along undisturbed tracts of land of rural areas.[xv] It is well established in the UK where it is found in a wide variety of manmade habitats. Widely perceived as a pernicious plant, knotweed occupies more than half of the area used to map plant distribution in the UK.[xvi] It is particularly abundant in riparian areas where it occurred along 84% of urban rivers with average flows > 2.3 m^3s^{-1}.[xvii]

2.1.4 *Ecological impacts*

In addition to its rapid spread, knotweed has caused many ecological problems in its introduced range. Its ecological impact on riparian ecosystems in the UK has raised serious concerns among plant ecologists. One the major impacts of knotweed is its ability to displace native plant species along riparian habitats. By forming dense stands along riverbanks, knotweed is able to restrict the growth of ground flora, causing a reduction in the diversity of native species.[xviii] Its dense and tall canopy can out-shade other plants. Even after knotweed dies back in the winter, its leaf litter prevents germination of native seeds.[xix]

As a result, the control of knotweed has been the focus of many ecologists in the UK who view it as a serious threat to species diversity and wildlife habitat. Considered one of the top 20 invasive aliens in Great Britain, it is illegal to plant knotweed in the UK under the Wildlife and Countryside Act (1981).[xx] Although there have been many attempts to eradicate the plant, the literature on eradicating established knotweed stands is not encouraging. In fact, every year the UK spends millions of dollars (14 pounds per square meter) to control the weed, mainly with chemical herbicides.[xxi]

2.1.5 *Status in United States*

Although knotweed has become increasingly common in North America, the species is not yet rated as a serious weed. In the US, Japanese knotweed is represented from the southern states to the northeast. In the western US, it is distributed from CA to the piedmont valleys of the Rocky Mountains.[xxii] Knotweed has not yet reached the level of repugnance as it has in the UK. It is listed on many plant lists such as the National Exotic Pest Plants List, the Eastern Region Invasive Plants List, and the Noxious Weed List of many states.[xxiii] However, there is no legal ban selling knotweed for horticulture and landscaping, planting or spreading the weed in the wild. Moreover, there is no specific policy on Knotweed eradication, control or management. In fact, many nurseries in the US and RI continue to sell the plant.[xxiv]

2.1.6 *Status in Rhode Island*

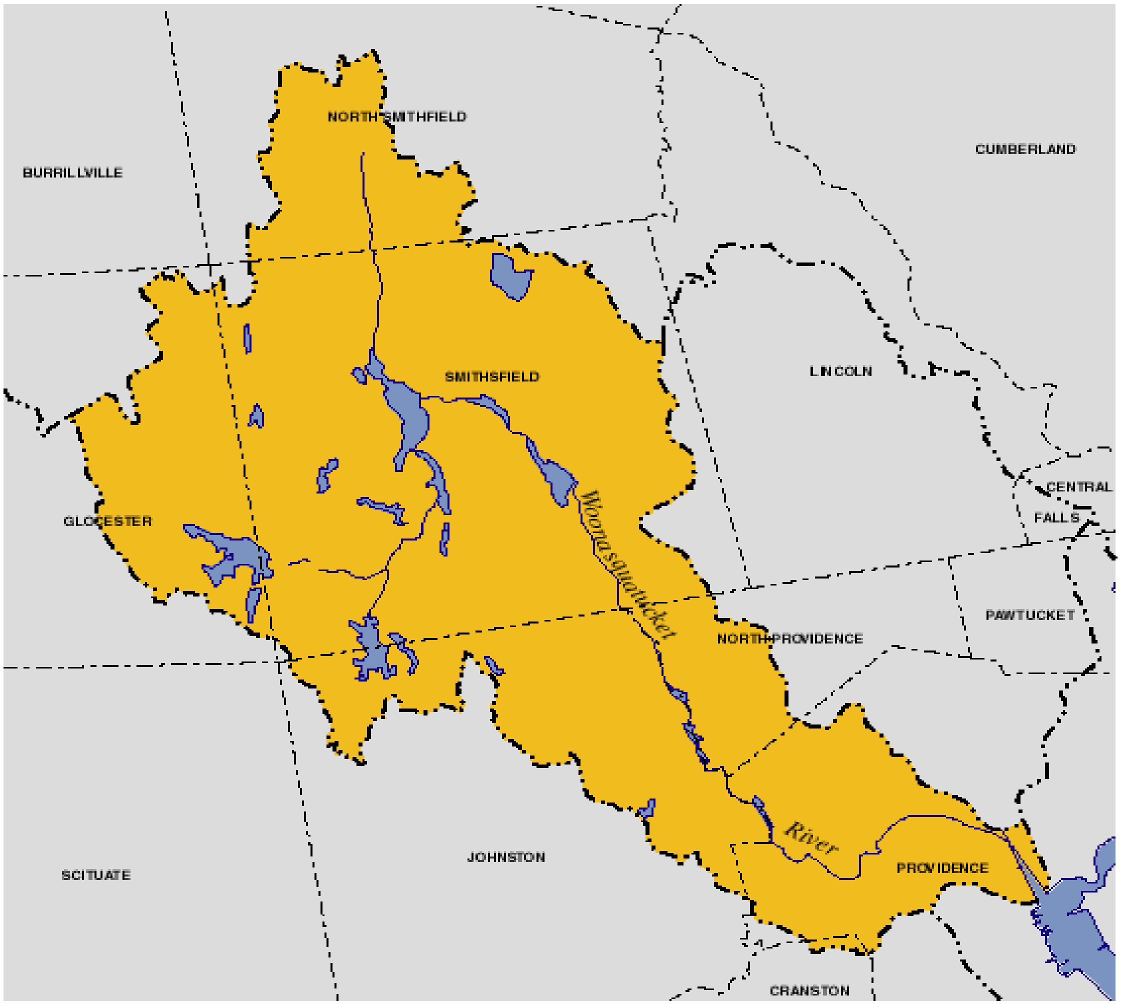
The legal status of Japanese knotweed in Rhode Island is similar to its national status. Although it is currently listed as an invasive plant, is not yet listed under the state's Noxious Weed List - [xxv] although Knotweed has been identified as a potential problem in many parts of the state by plant ecologists.[xxvi] However, its status has not yet reached that of Common Reed (*Phragmites australis*) and Purple Loosestrife **(***Lythrum salicaria***)**, two of RI's most invasive plants[xxvii]. Therefore, RIDEM does not have specific policies on its eradication, control or management. Nonetheless, Knotweed has caused problems to unmanaged vegetation of urban areas, and is seen everywhere from roadsides to residential backyards. Similar to the UK, knotweed is particularly invasive to riparian habitats, and is especially ubiquitous along disturbed stretches of the Woonasquatucket River.[xxviii]

2.2 ***The Woonasquatucket River***

2.2.1 History

The Woonasquatucket River runs the length of 18 miles, flowing through six cities and towns in Rhode Island including Glocester, North Smithfield, Smithfield, Johnston, North Providence, and Providence (Figure 2-1). [xxix] In Providence, it merges with the Mosshassuck River where it becomes the Providence River and flows out to Narragansett Bay. The upper section of the Woonasquatucket River Watershed (near Glocester and Smithfield) is relatively pristine and rural. The upper portion of the basin is characterized by agricultural use. In contrast, the lower portion of the river (bay-ward the Smithfield line) is characterized by commercial and industrial landuse, and has been polluted with sewage and industrial waste for more than a century.[xxx] Having played a significant role in the Industrial Revolution, the Woonasquatucket River was one of the first rivers to be dammed by mill owners and used for a source of water supply as well as a sink for chemical dumping. Located along the banks of the river, many chemical factories such as textile, paper and finishing industries used the Woonasquatucket River as their dumping ground for hazardous waste.[xxxi]

Figure 2-1. Map of the Woonasquatucket River Watershed



Even after many mills closed down and left the area, degradation of the river did not end. Currently, several industrial uses along the river continue to pollute the environment.[xxxii] Thus, for over a century, the river has been neglected and left in state of dereliction. As a result, it has gone from a valued asset to neglected natural resource. Water quality in the lower portion of the Woonasquatucket River has been, and continues to be severely impaired by inorganic contaminants such as heavy metals, dioxins, and PCBs.[xxxiii] In addition, much of the land adjacent to the lower portion of the Woonasquatucket River contains significant levels of heavy metal contamination.

2.2.2 *Revitalization and Restoration*

It was not until the Woonasquatucket River was designated an American Heritage River by congress in 1998, that significant effort was invested into restoring and revitalizing its banks and waters.[xxxiv] This designation enabled the river to receive federal assistance for up to 5 years in the form of refocused programs and technical assistance - Moreover, it served as a catalyst for attracting attention and funding towards improving the health of the river. Since its designation, many groups have taken an interest in revitalizing the river and the watershed that it runs through.

2.2.3 *Restoration Efforts*

Some of the major interested parties include the Environmental Protection Agency (EPA), Rhode Island

Department of Environmental Management (RIDEM), Narragansett Bay Commission (NBC), Woonasquatucket River Watershed Council (WRWC), city of Providence and other municipal governments, local watershed residents[xxxv]. Although the focus of these groups differs in terms of watershed restoration, their overall goal remains the same: to improve water quality in the river. The DEM and WRWC in particular are not only focused on maintaining water quality, but are also working to restore the riparian habitat adjacent to the river. Recognizing that the adjacent land also affects the health of the river, they have extended their revitalization efforts to the riparian buffers in the watershed. Using funds provided by the USDA Forest Service, the DEM and WRWC are working together to identify and evaluate sites along the river as potential sites for riparian buffer restoration[xxxvi]. The report, entitled the Woonasquatucket River Riparian Buffer Development Project (2001), which was used to identify these sites, also lists the major impacts on the riparian buffer. Some of these impacts include: (1) removal of forest vegetation, (2) impervious surfaces directly adjacent to river, (3) storm drains that bypass vegetated buffer areas, and (4) invasive exotic species, especially Japanese knotweed.[xxxvii]

2.2.4 *Japanese knotweed along the Woonasquatucket River*

The abundance and impact of Japanese knotweed has raised serious concern among restoration planners. Japanese Knotweed represents a significant component of plant assemblage in the watershed, and is ubiquitous along the lower parts of the riverbanks.[xxxviii] In order to restore the community of native species along the buffer, restoration planners recommend eradicating Japanese knotweed. However, is it wise to eradicate the species before knowing its value and what benefits it may provide? After all, eradication is both costly and labor intensive.[xxxix] The only way to successfully treat knotweed is through repeated applications of costly chemical herbicides such as glyphosphate.[xl] However, there are risks involved with using herbicides near watercourses. Mechanical removal, e.g. digging and uprooting the plant has not proven to be successful. As mentioned earlier, it is virtually impossible to eradicate knotweed at a reasonable cost.[xli] Thus, rather than focusing on the problems of knotweed, a more practical approach may be to consider the long-term benefits it may provide.

2.2.5 *Benefits of Japanese knotweed*

Some of the benefits of Japanese Knotweed include: its value as an ornamental and fodder plant, its use for erosion control, its medicinal attributes, its role as a primary successional species, its value as food source, and finally its role as a potential hyperaccumulator.[xlii]

2.2.6 *Japanese knotweed as a potential hyperaccumulator*

A hyperaccumulator is a plant that can tolerate and accumulate significant amounts of toxins without becoming phytotoxic.[xliii] These plants can tolerate exceedingly high amounts of toxins, at a level beyond which "normal" or non-hyperaccumulator plants can withstand. As a result, hyperaccumulators can thrive in soil contaminated to levels that are often orders of magnitudes higher than current regulatory standards.[xliv] In addition to being tolerant to harsh conditions, hyperaccumulators can also bioaccumulate toxins from the environment into their roots and shoots. In theory, hyperaccumulators can be grown and harvested economically, leaving the soil with a greatly reduced level of toxic contamination. Thus, if Japanese knotweed is determined to be a hyperaccumulator, it should be used to remove metals from contaminated soils along the Woonasquatucket River. Eradication is not a feasible option at this time.[xlv] For that reason, a long-term method to deal with knotweed invasion along the Woonasquatucket River must be developed. In addition to preventing its spread, the state should also take advantage of the value it may provide as a hyperaccumulator. Therefore, this study asks the question: Should the state and local authorities of Rhode Island implement a policy to manage Japanese knotweed along the Woonasquatucket River?

2.3 ***Phytoremediation and the response of Knotweed to metals***

2.3.1 *Phytoremediation defined*

Phytoremediation is the use of plants to remove or render harmless toxins in the environment.[xlvi] Since the early 1990s, this new and emerging technology has become a widely considered method for cleaning up toxic sites. Phytoremediation has many advantages. First, it is cost effective. The cost of remediating soils (conventionally) is much greater than the costs of planting and removing hyperaccumulator plants.[xlvii] Conventional methods of remediating soil often include excavation of site and chemical treatment of soil. Secondly, it is less destructive. Instead of removing toxins from a site using ex-situ techniques, hyperaccumulator plants can be removed from sites without disturbing neighboring vegetation. Finally, harvested metals from plants can be recovered and recycled from collected burnt ash.[xlviii] A subset of phytoremediation is phytoextraction, which is a process in which “metal-accumulating plants are used to transport and concentrate metals from soil into harvestable parts of roots and above-ground shoots.”[xlix]

2.3.2 *Criteria for a hyperaccumulator*

In order to evaluate knotweed’s potential in phytoremediation, this study focuses on comparing the Jk’s metal accumulating abilities to that of a hyperaccumulator plant. The criterion for hyperaccumulation represents a concentration in aboveground dry matter in excess of ‘normal’ or physiological levels. Since there is no clear distinction between normal and excess concentration, many studies have applied their own criteria for a hyperaccumulator. Some definitions include (1) Most hyperaccumulators can accumulate up to 1000 ug g^{-1} of lead and copper, and up to 10,000ug g^{-1} of zinc in their tissues.[l] (2) Hyperaccumulators accumulate large amounts of metal irrespective of metal concentration in soil. (3) Hyperaccumulators show a ratio greater than 1 for concentrations of metals in root vs. shoot tissues.[li] These criteria will be addressed when comparing the ability of knotweed to bioaccummulate heavy metals to known hyperaccumulators.

2.3.3 *Plant response models*

To understand the response of knotweed to metal contamination, this study examined three basic strategies that plants employ for growing on metalliferous soils: metal excluders, hyperaccumulators and indicators.[lii] Metal excluders can contain large amounts of metals in their roots, but effectively prevent metal from entering their aerial parts over a broad range of soil concentrations. Hyperaccumulators, on the other hand, can accumulate large amounts of metals in their roots irrespective of soil concentration. In contrast, metal levels in indicator species generally reflect metal in soil.

2.3.4 *What makes Japanese knotweed a good candidate?*

One of the major challenges that scientists in phytoremediation face is the selection of a “good” phytoremediator for a particular site. Only a handful of hyperaccumulators have been identified for use in phytoremediation. The members of the Brassicaceae family have been extensively researched for their ability to bioaccumulate heavy metals.[liii] Although they are able to hyperaccumulate high amounts of toxins, such as nickel, their small size and biomass make them less effective at decontaminating sites in a reasonable number of harvests. For example, the shoots of *Thlaspi rotundifolium* reportedly contain up to 8200 mg Pb kg^{-1} dry wt, but these plants amass only up to 50 mg/plant of dry tissue months of growth.[liv] Therefore finding a plant that has most if not all the attributes of a “good” phytoremediator becomes a difficult task. Some of the criteria used to select plants for phytoremediation include: rapid growth rate, large biomass, deep and extensive roots, and tolerance of harsh environments.[lv] To compare, Japanese knotweed can grow up to 4.65 cm/day during the height of its growing season (March- July),[lvi] weighs on the average 101.4g/plant dry wt,[lvii] has extensive roots that extend down to 2m,[lviii] and is tolerant of environments with low pH, high salinity, and high heavy metal concentrations.[lix] More importantly, it is firmly established along the urbanized banks of the Woonasquatucket River. Having to plant hyperaccumluator plants is costly, time consuming, and labor intensive. In addition, a lot of maintenance is required to take care of the plants. Japanese knotweed is both invasive and abundant along the river, therefore harvesting the plant might serve two purposes: control of an invasive, and remediation of contaminated soil. Therefore, Japanese knotweed makes good a candidate to examine in this study.

3. **Methods and Materials**

Data collection and sample preparation was assisted by Professor Jim Baird’s Environmental Chemistry course.

3.1 ***Site Selection***

Prior to sample collection, various sites along the Woonasquatucket River were identified and physically surveyed with assistance from the students in Chemistry 117. The sites were selected primarily based on the Woonasquatucket River Riparian Buffer Development Project Draft Report 2000, which had evaluated sites along the Woonasquatucket River for potential restoration. Selected sites were labeled according to the report’s definition for consistency, with minor adjustments in location of a few sites. The criteria used for site selection for this study included: presence and abundance of Japanese Knotweed patches, tract length, accessibility of site for sampling, and proximity of site to pollution sources (e.g. sewage drains and adjacent mills), relative distance between sites. Thirteen sites along the Woonasquatucket River were chosen for sampling (Figure 3-1). Of the 13 sites chosen, seven were novel to the presence of Japanese Knotweed. These sites are located in the lower portion of the watershed, in Johnston and Providence. The remaining six

Figure 3-1. Map of sampling sites along the Woonasquatucket River (Appendix B).

sites, which are located in the upper portion of the watershed (Glocester and Smithfield), were chosen to help map gradients in concentrations of metals along the river. Sites located in the upper portion of the watershed are devoid of Knotweed. All 13 sites are located approximately 1 mile from each other, and are grouped in four clusters, which represent sites from each township. Clusters 1 and 2 represent sites with abundant Japanese Knotweed. The following is a table of all sites, with their approximate location in the watershed:

Table 3-1

Site	**Street Location**
Cluster 1	Providence
P2	Acorn bridge St. off Promenade St. and past Pleasant Valley
P8*	Valley St. Park, past Atwells
P10*	Valley and San Souci (near Price Rite)
P13	Merino Park
Cluster 2	Johnston
J01	Intersection of Route 44 and 128 (Putnam Pike)
J05*	Route 104 and Warren Ave.
J02	Greystone Ave. and Angell St. (bridge off 104)
Cluster 3	Smithfield
S11	Whipple Field
S05	Smith Appleby (Stillwater Rd.)
S04	Stillwater Mill (Stillwater Rd. Thurber Ave.)
Cluster 4	North Smithfield/Glocester
S03	3 Spragueville Rd.
S06	Route 44/Greenville Rd.
G02	Glocester Country Club

*indicates sites that are very close in proximity, but not in the exact location of Kleinschmidt sites.

3.2 *Sample collection*

All samples were collected in late September to early October of 2001. Sediment samples were taken from all sites, while plant and soil samples were only taken from the 7 sites with presence of Japanese knotweed (Figure 3-2). Sediment samples were collected from the bottom of the river using a plastic scoop. Samples were collected in polypropylene tubes and stored at 4° C until ready for digestion. Where 1-3 (circular monoclonal stands) patches of knotweed was found, transects of 30m-70m were measured encompassing all patches for each site. The patches of Japanese Knotweed were labeled with numbers 1-3, starting with patch furthest upstream. While the majority of Japanese Knotweed sites had three patches, there were a few sites with less than 3 patches. Site J05 and all sites in Cluster 1 had 3 patches of Japanese Knotweed, while 2 patches were found in J02, and 1 patch in J01. Plants of uniform size (large), condition (healthy), and proximity (directly adjacent to water) to the river were chosen for sampling. Three replicate plants were taken and dug out of the soil from each patch. Representative samples of the entire plant, e.g. leaves, shoots and roots were collected from each replicate and composited to per patch. Approximately 10 leaves and 1-2 ft long segments of shoots,

Figure 3-2. Sample Matrix - outlines how samples were collected from each site (Appendix A).

and root pieces from each plant were removed and placed in labeled plastic Ziploc bags. In addition, soil samples were collected in the immediate vicinity of roots of knotweed plants that were collected. To maintain high quality control of the plant tissue analysis, special precaution was taken to avoid various sources of potential contamination. Plastic gloves were used when collecting and handling samples, and storage and transportation of samples were conducted using non-metallic materials.

3.3 ***Site Description***

Site conditions, physical characteristics and identity of associated plants were recorded in addition to sample collection. (Appendix A). The majority of plant species were identified at each site with the assistance of Eugenia Marks of the RI Audubon Society. For each transect of bank, the linear proportion of the bank occupied by the species (percent cover of knotweed) was estimated visually as an expression of abundance. Dates of collection and specific data are available in Appendix A.

3.4 ***Sample Preparation and Organic Digestion***

Upon collection, all samples were immediately brought back to the lab where they weighed, carefully cleaned and wiped, then oven-dried (Lindberg/Blue Laboratory Drying Oven) at 80°C for 48 hrs. Special care was given to roots samples to minimize soil contamination. The roots were cleaned with absorbent paper and carefully removed of soil particles. The roots were then cut into small pieces and homogenized by plastic tweezers. To detect soil contamination washed roots were compared to unwashed roots for significant differences in metal concentration. Excess plant material and debris were filtered from the soil samples after drying. After all plant and soil samples were dried, a small representative sample from each bag was homogenized using plastic tweezers. Trace element analyses were carried out by acid digestion of dried and ground tissues. Approximately 0.05g was digested in concentrated 70% HNO3 at 180 degrees (Ethos 1600 Advanced Microwave Labstation- Milestone Microwave Laboratory Systems), according to the standard operating procedures. (Appendix B). Digestions were carried out with minor adjustments in sample weight and use of Teflon inserts. As part of quality control, National Institute of Standards and Technology plant (peach and pine leaf) reference standards were carried through the digestion protocol and heavy metal analysis. A comparison of the results of this study with the published values for these samples is given in Appendix B. Reagent blanks were also used where appropriate to ensure accuracy and precision in analysis.

3.5 ***Heavy Metal Analysis***

After digestion in nitric acid, plants were analyzed for selected metals by Perkin Elmer Model 4100ZL Zeeman Atomic Absorption Spectrometer (AA) and Jobin Yvon Horiba Model JY 2000 Inductively Coupled Plasma Spectrometer (ICP). The elements chosen for compositional analysis were Lead, Copper, Chromium, Cadmium, Nickel and Zinc. The heavy metals chosen are ubiquitous pollutants present in industrial, agricultural, and municipal waste. Only “available” or soluble metals were analyzed in all samples. Lead, Copper and Chromium were measured by the AA, while Cadmium, Nickel and Zinc were measured by ICP. Samples analyzed for lead composition were run on both machines to ensure compatibility of results. For the AA, instrument readings were verified daily by running Certified (NIST) standards of a known composition (Appendix B). Standard solutions were prepared for analysis on the AA and ICP from Certified Standards for atomic absorption spectrophotometry and appropriate amounts of 70% concentrated acid was used for dilutions. The AA and ICP were operated according to the standard procedures given by Mr. Joseph Orchardo (Appendix B). The sensitivity ranges for the machines are ppm and ppb.

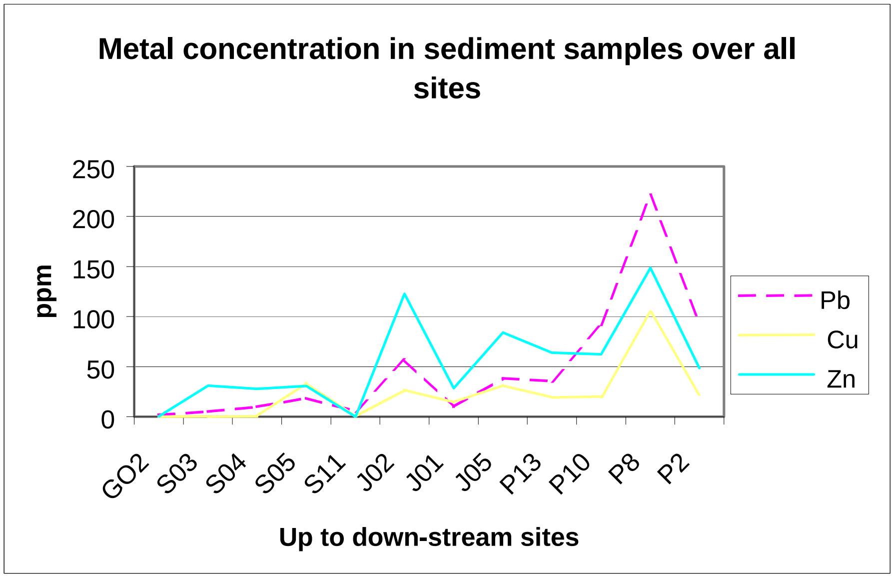
4. **Results**

Before any data was analyzed, a statistical test to determine whether there were any significant differences in metal concentration of samples between patches in all Japanese knotweed sites was run. Results of the one-way ANOVA showed that there were no significant differences between patches (P>0.05). Therefore, the values from each patch were used as replicates for each site.

4.1 ***Analysis of sediment and soil samples***

Metal concentration in sediment samples increased from upstream sites (Glocester and Smithfield, sites G02, S06, S03, S04, S05, S11) to downstream sites (Johnston and Providence, sites J02, J01, J05, P13, P10, P8, P2). Out of the six elements measured in sediment samples, only lead, copper, and zinc were above detection limits. Figure 4-1 shows a general gradient of metal concentration along the river, with the highest concentration of lead, zinc and copper in sites J02 and P8.

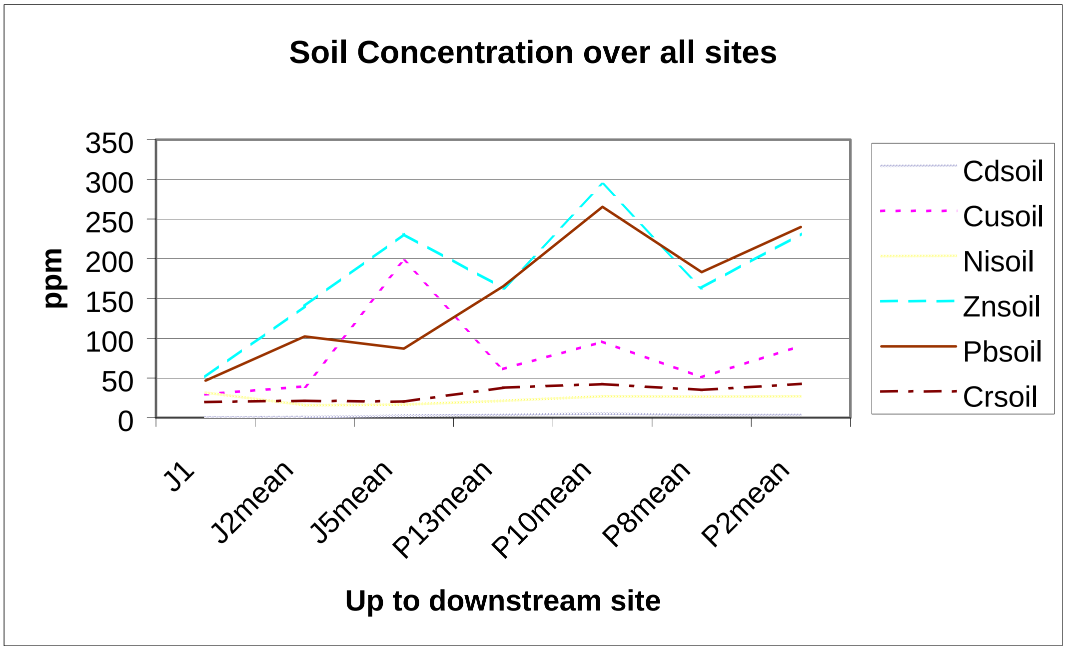
Figure 4-1



Concentration (ppm) of Lead, Zinc and Copper in sediment samples collected from all sites (upstream to downstream).

Nickel, cadmium and chromium concentrations were below the detection limits for the AA and ICP at all stations for all samples types. Therefore, they are not represented in Figure 4-1. The most prevalent metals (in descending order) in the soil samples are zinc, lead and copper (Figure 4-2). Zinc concentration over all sites ranged from 500-85ppm, while lead and copper concentration ranged from 265-48ppm, and 198-30ppm. Similar to sediment samples, nickel, cadmium and chromium concentrations were significantly lower than zinc, lead and copper concentrations. In addition, all plant parts of knotweed accumulated significantly more zinc, lead and copper than nickel, cadmium and chromium (see next section). Thus, for the remainder of this study, only lead, zinc and copper will be included in future analyses. The most contaminated site, P10 had the highest concentration of lead and zinc, while J5 had the highest concentration of copper.

Figure 4-2



Mean metal concentration (ppm) of soil samples collected from sites with Japanese knotweed along the Woonasquatucket River.

The only metal that exceeded the state’s regulatory limits for direct exposure of metals in residential soil was lead at site P10. However all metal concentration in all sites exceeded the expected background levels of pollutant metals in RI soils (Table 4-1).

Table 4-1

Element	**RI Direct Exposure Criteria (Residential) (ppm)**	**Background levels in RI (ppm)**	**Soils along Woonasquatucket River (P10) (ppm)**
Cadmium	39	0.22-3.5	5
Chromium IV	390	9	42
Copper	3100	14	96
Lead	150	34	265
Nickel	1000	7	27
Zinc	6000	42	293

Comparison of metal concentration in soil samples collected from the most contaminated site (P10) to Residential Direct Exposure Criteria for metals in Rhode Island soils, and Background Levels of Priority Pollutant Metals in Rhode Island Soils.

4.2 ***Response to metal in plant parts of Japanese knotweed***

The response of plant tissue to metals differs considerably; therefore, each metal will be analyzed individually. Figure 4-3 shows the distribution of zinc, lead and copper (total metal) in the roots shoots and leaves of Japanese knotweed along the Woonasquatucket River. Distribution in the plant parts was not even, but was consistent for all metals. In all sites, the highest metal content was found in roots, followed by leaves, then shoots. Mean zinc, lead and copper concentrations were found to be highest in the roots, and evenly distributed in leaves and shoots. Figure 4-4 represents the mean concentration of metal distributed in the different parts of the plant.

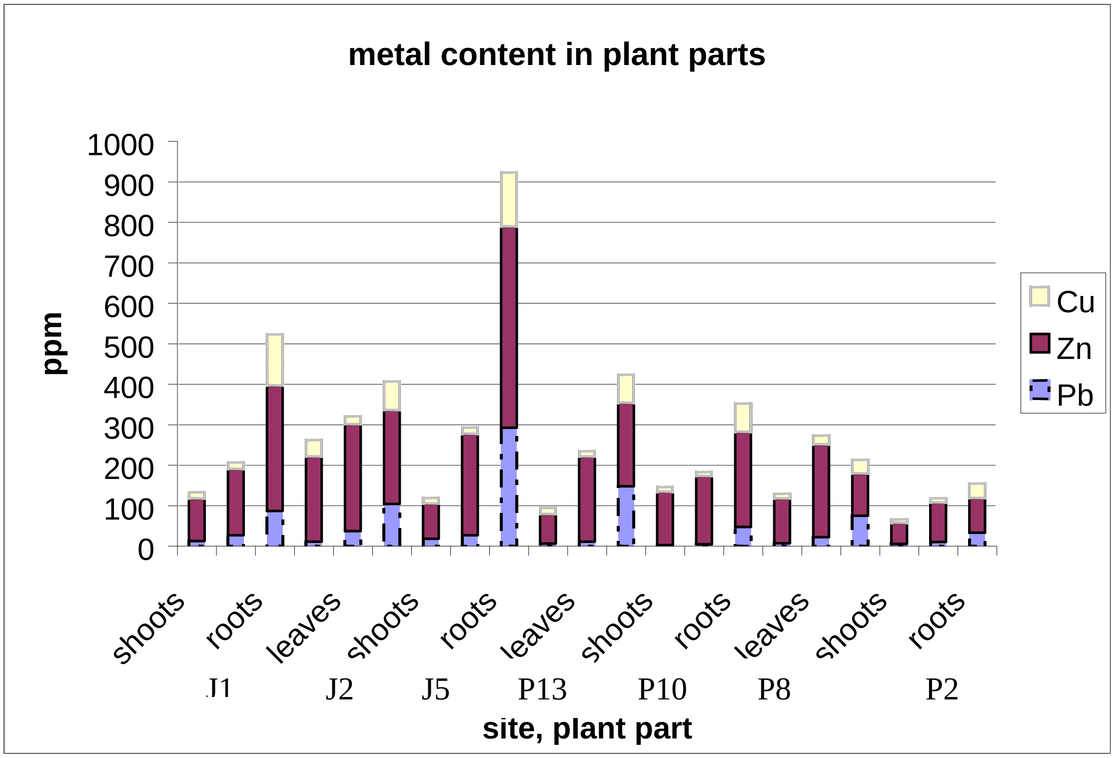


Figure 4.3. Metal Concentration in leaves, roots, and shoots of Japanese knotweed in all plant sites.

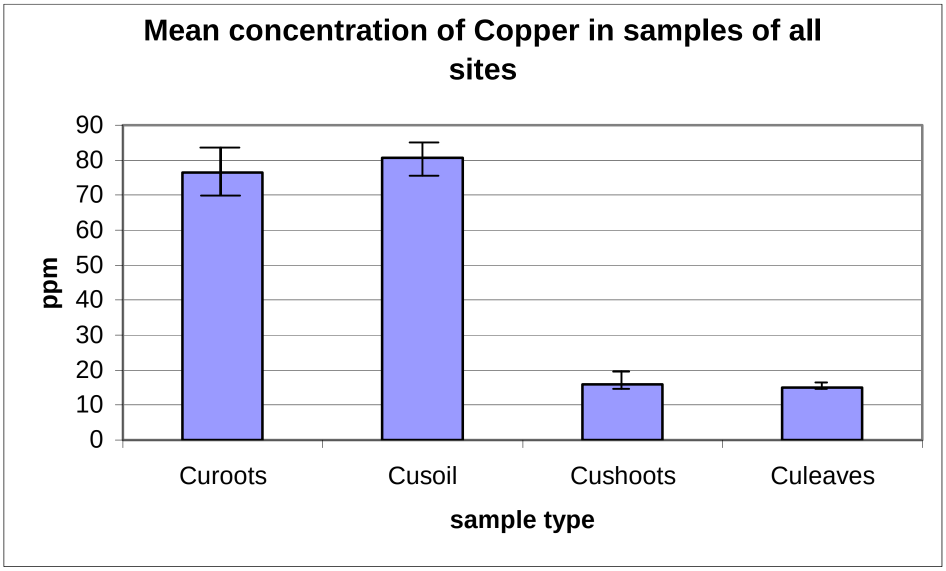


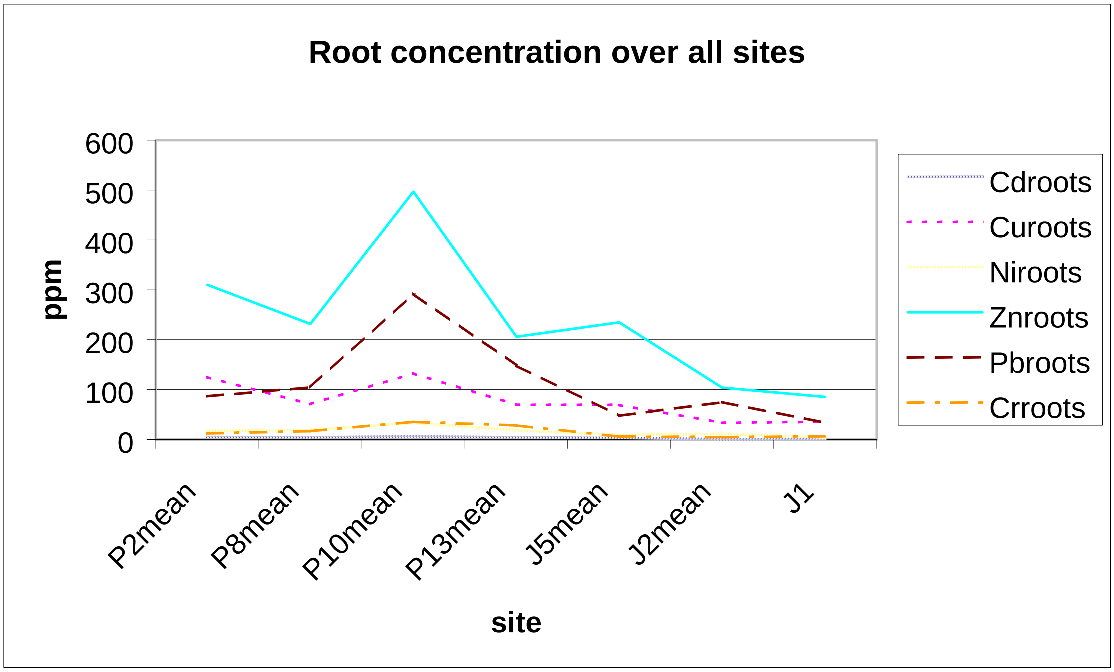
Figure 4-4 Mean concentration of copper in plant parts.

The roots in site P10 have the highest amount of all three metals, reflecting zinc, lead and copper concentration in soil for site P10 as well (Figure 4-5). Zinc concentration in roots ranged from 500-85ppm, while lead and copper concentration ranged from 293-33ppm, and 133-33ppm. Based on the published biomass of knotweed shoot (101.14 g), percent dry weight of zinc in all plant parts was less than 0.01%. Compared to soil, the concentration of zinc was much greater in the roots for some sites. Shoots and leaves from all sites had the highest concentration of zinc, reaching as high as 200ppm. Lead and copper on the other hand were comparatively low in the aerial parts of the plant (Appendix C).

4.3 ***Metal concentration in roots***

Zinc and lead concentration in soil correlates with concentrations in roots of Japanese knotweed. Figures C1 and C2 of Appendix C show a linear relationship between soil and root concentration where mean concentration on root increased with the external (soil). The increase in root concentration is explained by an increase in soil concentration for both zinc ($R^2 = 0.8130$, $P<0.05$) and lead ($R^2 = 0.5884$, $P<0.05$) concentrations.

Figure 4-5



Metal concentration (ppm) in root samples of knotweed sites.

There was no linear relationship between root and soil concentration for copper.

4.4 ***Metal concentration in shoots***

Metal concentration in shoots did not reflect increasing soil concentration as well (Appendix C). Copper and zinc concentration in shoots ranged from high to low concentrations over the different sites with no particular trend. Unlike the correlations seen for copper and zinc, lead concentration in shoot was low and constant for all sites irrespective of soil concentration.

Although metal concentration in shoots from all sites were significantly lower than the corresponding roots, the shoots from sites P2, P8, and P10 are comparatively higher than the expected “normal “ abundance of vegetation for zinc, lead and copper. “Normal” abundance refers to the average amount of metal found in vegetation growing in non-metalliferous soil. Table 4-2 shows the mean concentration of zinc in shoots from site P8 is more than 100% greater compared to the normal abundance of zinc found in vegetation.

Table 4-2

Metal	**Normal abundance in vegeta-tion**	**Site P2**	**Site P8**	**Site P10**	**Site P13**	**Site J5**	**Site J2**	**Site J1**
Lead	5	16	18	20	7	3	11	6
Copper	10	15	35	13	14	11	13	9
Zinc	100	120	224	128	107	141	141	64

Comparison of mean zinc, lead, copper in aboveground biomass of Japanese Knotweed, at a range of sites along the Woonasquatucket River, to normal abundances in found in worldwide vegetation. Concentrations are in ug g^{-1} dry wt.
Source: Brooks, R.R. (1987)

4.5 ***Comparison of roots to shoots***

The ratio of aerial biomass (shoots and leaves) to roots was found to be less than 1 for concentrations of lead, copper and zinc at all sites. With the exception of zinc concentration in plant samples collected from all sites, the ratios calculated for copper and lead concentration were less than 0.5 (Appendix C). Zinc, lead and copper concentration in roots were found to be 2-3 times greater than shoots.

Using the published biomass of knotweed, in addition to patch area (m^2) at each site, the amount of each metal removed by harvesting shoots from soil (on an annual basis) was calculated for each site. Table 4-3 compares the amount of metal removed from each site. Sample calculations are available in Appendix C. Harvesting shoots from Site P2 and P8 would remove the greatest amount of zinc, copper and lead. Harvesting from site J1, on the other hand would barely remove any zinc, copper or lead.

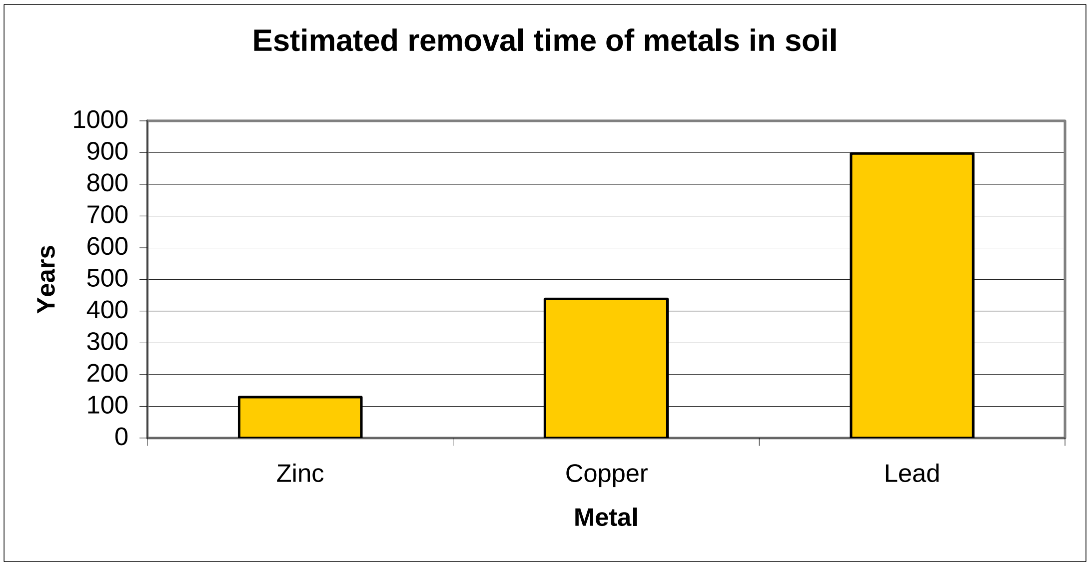
Table 4-3

Site	**Cu (g/site)**	**Zn (g/site)**	**Pb (g/site)**
P2	3.29	25.76	3.49
P8	6.55	42.07	3.36
P10	1.05	10	1.58
P13	1.37	10.4	0.72
J5	0.78	10.35	0.23
J2	0.48	5.08	0.39
J1	0.089	0.65	0.065

Amount of metal removed from each site. Calculations are based on mean metal concentration in shoots from patches of each site.

The number of annual harvests required to reduce metal concentration in soil to acceptable levels (i.e. background levels) was calculated for the most contaminated site, P10. Two assumptions were made prior to these calculations: (1) subsequent patches of knotweed would remove metals at the same rate as the first crop, and (2) the rate of metal deposition is neglible. Thus, if there are 75,000g[lx] of soil in a cubic meter, the number of years it would take to remove copper, zinc and lead from site P10 was calculated to be 438, 129 and 897 years (Figure 4-6).

Figure 4-6



Metal removal was calculated for the most contaminated site, P10. Calculations are based on assumption that a cubic meter of soil weighs approximately 75,000g.

4.6 ***Plant survey***

Only sites in the lower portion of the watershed were abundant with Japanese knotweed. No other native plant was found to co-occur with knotweed in these sites. Many other exotic plants, such as Tree of Heaven and Asian Bittersweet, were also present in these sites (Appendix A). However, there was no particular trend in the ratio of natives to exotics with respect to knotweed abundance or soil contamination. The most common co-occurring exotic plant species found with Japanese knotweed was Tree of Heaven (*Ailanthus altissima)*. Its abundance was comparatively lower than knotweed abundance though. Percent cover of Japanese knotweed observed at each site did not correspond with elevated metal concentration in soil (Appendix C). Sites with most percent cover of knotweed were not necessarily the most contaminated soil sites. However, the site (J05) with the greatest abundance of knotweed also corresponded with the site with the highest concentration of zinc and copper.

5. **Discussion**

5.1 ***Heavy Metals in Sediment and Soils***

As expected, the gradient of increasing metal concentration (moving downstream to Providence) confirms the fact that the lower portion of the watershed is more impacted by industrial pollution than the upper portion. Additionally, the fact that knotweed is not present in sites above the Smithfield line supports the knowledge that knotweed is particularly invasive to derelict and disturbed sites in urban areas. [lxi] The lower portion of the watershed is characterized by urban development and industrial landuse, therefore it is not surprising that knotweed has become abundant along the banks of this section. Although soil levels in this study do not exceed state regulatory limits, they are higher than background levels for pollutant metals in Rhode Island. It is not surprising that the soils are moderately contaminated with heavy metals. Metal concentrations in soils were expected to be higher based on proximity to pollution sources such as industrial mills and sewage drains.

5.2 ***Metal tolerance in plants***

Metal toxicity in plants can cause leaf scorching, nutrient deficiency, shortening of roots and increased vulnerability to insect attack[lxii]. However, metals do not appear to be phytotoxic to knotweed. The knotweed plants in this study accumulate measurable zinc, copper, and lead without apparent physical damage or physiological strain. Raskin suggests that the phytotoxicity of heavy metals like cadmium and copper is substantially greater than lead and zinc.[lxiii] This may explain why zinc concentration is substantially higher than copper in aboveground tissue. Zinc is an essential micronutrient and plays an important role in the both carbohydrate metabolism and protein synthesis.[lxiv] Its solubility makes it easily translocated to different parts of the plant. The results of this study show that the roots of knotweed are able to translocate high amounts of zinc to its shoots. Brooks suggests that plants can take up large quantities of zinc without evidence of exclusion when Zn concentrations in soil are less than 500mg/kg.[lxv]

Copper is also an essential element to plants. However, only large quantities of copper were located in the roots of knotweed. In fact, very little copper was translocated to shoots from roots. Other studies have shown that roots with high quantities of copper do not effectively translocate the element to shoots. Jarvis et al. point out that plants growing in high copper environments often translocate surprisingly little copper to their aboveground biomass but generally accumulate large amounts in their roots.[lxvi] Kabata-Pendias et al. observed that under conditions of both Cu deficiency and excess, root tissues strongly hold on to Cu against the transport to shoots.[lxvii]

Unlike zinc and copper, lead is not considered an essential element to plants, and only a small proportion of lead is bioavailable to plants.[lxviii] The roots of knotweed were considerably higher in lead concentration than the shoots or leaves. Thus, there is little translocation of lead from roots to shoots. Although lead has a very limited availability, plant roots are usually able to take up and accumulate large quantities of lead in soil.[lxix] However, translocation of lead to aerial shoots is generally limited due to binding to root surfaces and cell walls.[lxx]

Although Japanese knotweed has been observed to tolerate elevated concentrations of heavy metals, it does not accumulate great amounts of lead, copper or zinc in aboveground biomass. This is supported by the results of Kubota et al, which shows copper tolerance in knotweed roots.[lxxi] In a previous study by Kubota et al, large amounts of Cu, Zn, and Cd were concentrated in the roots of

Japanese knotweed growing in soils contaminated with Cu, Zn, or Cd.[lxxii]

5.3 ***Comparison to known Hyperaccumulators***

Hyperaccumulator species have the ability to accumulate large amounts of contaminants in both shoots and roots. Although it is able to concentrate metals in its above and belowground tissues, the metal levels found in Japanese knotweed shoots do not exceed those present in the soil. Recognized hyperaccumulators like *Thlaspi caerulescens* have been shown to have shoot concentrations ranging from 13000 to 21000 mg kg-1 (dry weight) growing in contaminated sites in the British Isles.[lxxiii] In comparison, metal concentration in roots of knotweed show that it does not accumulate up to the level of established hyperaccumulators. In addition to accumulating high amounts of metal in its roots, a hyperaccumulator plant must also have the ability to translocate the element from roots to shoots at high rates.[lxxiv] Kufel defines hyperaccumulators as plants which show a ratio greater than 1 for concentrations of metals in root vs. shoot tissues.[lxxv] On the contrary, root to shoot in Japanese knotweed showed ratios less than 1. One possibility is that the roots may be accumulating large amounts of metals, but are not efficiently translocating the metals to shoots.

5.4 ***Plant response models***

The three basic strategies that plants employ for growing on metalliferous soils have been discussed in a previous section. Knotweed may not be a hyperaccumulator, but it does accumulate more metals than the average plant. If it cannot be categorized as a hyperaccumulator, what model of plant response does it best fit? The majority of plants that occur on metalliferous soils are known to exclude toxic metals from their shoots.[lxxvi] Knotweed behaves like a metal excluder in that it accumulates more zinc, copper and lead in its roots than its shoots. This fact suggests that translocation of metals from roots to shoots could be a limiting factor for the bioconcentration of these elements. However, analysis of shoot concentration does not entirely confirm patterns of exclusion. Shoot concentration of plants from most sites are higher than expected normal abundances, suggesting that the roots are not limiting translocation of metals to shoots. The results of this study show a linear relationship between soil and root concentration of zinc and lead. Metal levels in indicator species generally reflect metal in soil. Antonovics et al suggest that shoot to root ratios of metals are less than one when “indicator” species are grown on contaminated soils.[lxxvii] Therefore, the response of knotweed to metals is that of a metal indicator.

5.5 ***Metal Removal***

The roots of Japanese knotweed accumulate the most metal; therefore it is best to harvest roots for phytoremediation of soils. However, one time removal of knotweed will not achieve long-term phytoremediation goals. Harvesting the plant non-destructively allows the roots to accumulate metals overtime, thereby facilitating long-term removal. Therefore, one question remains to be answered: How long will it take to remediate metals in soil if shoots and leaves, i.e. aboveground biomass, are harvested instead? The results of this study show that it would take over 100 years of annual knotweed cropping to remove zinc, copper and lead from soils along the river. The removal rate of zinc is considerably lower than that of lead and copper. However, the time it would take to reduce the zinc levels in the soil is extremely long and impractical. Brown et al predicted that annual croppings on the order of 20 years would result in near total removal of target heavy metals.[lxxviii] Additionally, recent research suggests that "phytoremediation can deliver significant benefits in upland settings over a timeframe of under twenty years."[lxxix]

The number of years predicted for knotweed suggests that extraction of metals under harvesting conditions is not a realistic and economically viable option to remediating superficially contaminated soils along the river. Thus, there is little potential for the use of Japanese knotweed for metal removal along the Woonasquatucket River. Based on criteria used for phytoremediation, the results of this study show that Knotweed is not effective at remediating Zn, Pb, and Cu from contaminated soils.

6. **Conclusions and Recommendations**

6.1 ***Summary***

This study examines the value of Japanese knotweed in phytoremediation of contaminated soils along the Woonasquatucket River. Japanese knotweed is ubiquitous along the disturbed portions of the Woonasquatucket River.[lxxx] It is firmly established in areas that have received metals wastes from historic industrial discharges.[lxxxi] Although knotweed is not listed under Rhode Island’s Noxious Weed list[lxxxii], it is considered a serious threat to native plant species along the riparian buffer.[lxxxiii]

According to the EPA’s criteria for a good phytoremediator, Japanese knotweed makes a good candidate to evaluate in this study.[lxxxiv] Japanese knotweed bioaccumulates copper zinc and lead in its roots, shoots and leaves. The greatest amounts of metals are found in roots of knotweed. Copper, zinc and lead concentrations in roots correlate to corresponding metal concentrations in soil. However, abundance of Japanese knotweed is not correlated to sites with the highest soil concentrations of Cu, Zn, and Pb. Thus, Knotweed is tolerant of soils moderately contaminated with zinc, copper and lead. Furthermore, it is not a hyperaccumulator, and is not effective at remediating toxic soils along the Woonasquatucket River within a reasonable timeframe. Therefore, it should not be protected based on its phytoremediation properties.

Although many Asian countries have benefited from using knotweed as a source of food and medicine, the plant has very little value in Rhode Island. A benefit of knotweed that might contribute to its management is the prevention of soil erosion. However, the benefits of using knotweed for erosion control do not outweigh the costs of losing native plant species. Therefore, immediate action is necessary in order to prevent the spread of the species to other parts of the state. A policy to control and manage the plant should be implemented before it becomes a threat to ecosystems in Rhode Island.

6.2 ***Knotweed prevention and control***

There is no quick and easy way to control knotweed. The best method of control is preventing the species from becoming established. In order to do this; the RI Natural Heritage Program should include Japanese knotweed on its list of noxious weeds. In addition, the DEM’s Division of Agriculture should regulate the sale of JK at RI nurseries. To prevent nurseries from selling different cultivars of the plant the Division of Agriculture should also prohibit the sale of knotweed cultivars under the following scientific names: *Polygonum cuspidatum, Reynoutria japonica*, and *Fallopia japonica*.

Once it has established, the possibility of uprooting Japanese knotweed is very unlikely. Moreover, digging up knotweed roots is not suggested because it can lead to stranded root fragments that can regenerate and repopulate the area.[lxxxv] As mentioned before, Japanese knotweed is extremely difficult to eradicate by mechanical means. However, JK can be eliminated by repeatedly cutting and removing the stalks.[lxxxvi] Continued harvesting of shoots will not only exhaust the rhizome, but also allow more metals to be removed over time. While the aerial shoots of knotweed survive for one growing season, the rhizomes remain alive for more than a decade.[lxxxvii] Therefore, regular cutting after a number of years will eventually exhaust the rhizome and kill the plant. Since knotweed grows vigorously during the months of May-July, it is recommended that annual harvests occur in late July.[lxxxviii] Selection of removal sites should correspond with sites most contaminated with zinc, lead and copper since knotweed appears to concentrate these metals. Removing aboveground biomass of knotweed would also remove metals accumulated in the plant. Thus, sites that are both abundant with knotweed and highly contaminated with heavy metals should be targeted for removal. These sites are P8 and P10. It is also necessary to harvest the shoots so that the metals accumulated in plant shoots do not return to the environment by leaching or decomposition of the shoots. Annual removal should be sufficient at this time. In addition, the costs of handling, processing and subsequent land filling costs should also be considered. The amount of metal accumulated in knotweed is not high enough be considered as hazardous waste. Therefore, knotweed shoots could be composted and disposed of in a licensed landfill.

6.3 ***Education and public involvement***

It is important to keep the public informed and involved in the management of Japanese knotweed. Its removal can be used as a way to attract funding as well as to connect people to the river. For example, removing knotweed from the most contaminated sites will engage watershed residents (who are primarily concerned with water quality) in other activities that such as habitat restoration. In addition, knotweed removal can also be used to raise public awareness of issues concerning the reinvasion and spread of the plant. Proper identification and treatment of knotweed continues to be a problem in the state. In an attempt to eradicate knotweed on their properties, watershed residents are unknowingly spreading the plant by digging up the rhizome. Therefore, knotweed control should be the focus of watershed organizations such as the WRWC and Providence Plan. More importantly, effective management of knotweed requires a community-based effort. Thus, a policy to manage knotweed should encompass control, include the provision of information about the plant, raise awareness towards preventing reinvasion, and attract funding for long-term removal.

6.4 ***Restoration of native plants***

Targeted sites must be managed continuously until knotweed is eliminated and other plants are established. In order to allow for the growth of other plants, site clearance in the winter is also essential. It is also important to monitor the change in vegetation at sites where knotweed is controlled. There are two questions that should be addressed before the replanting of natives is considered: (1) Can native plants survive in polluted soils, (2) After knotweed is removed, what other plant will take its place? According to the plant survey of knotweed sites, no co-occurring native plant was observed where knotweed was abundant. This could be explained by the fact that these sites were also abundant with other exotic plants. However, there is no way of knowing what factor is limiting the presence of native plants in sites that are highly contaminated. The most common exotic plant that was observed to co-occur with knotweed is Tree of Heaven. This species is also known to be tolerant of polluted sites, but is not as menacing as knotweed along the river.[lxxxix] With so many variables to consider, it is difficult to speculate what changes will occur in the vegetation once knotweed is removed. This topic will require future work. Nevertheless, replanting of natives should not occur until these questions are properly addressed.

6.5 ***Future Research***

This study raises many questions that can only be answered by conducting additional research. The questions that are particularly interesting to parties involved in the restoration of the watershed are: (1) Are there any native plants in the watershed that can be used to remediate toxic soils along the Woonasquatucket River? (2) How will knotweed removal affect the plant assemblage in each site? (3) How long will it take to completely restore the community of native plants along the riparian buffer of the watershed? These questions are difficult if not impossible to answer at this stage. However, continued research that is built upon this study may shed some light in the near future. The fact is Japanese Knotweed is here, and will not go away unless action is taken to control its spread. Therefore, it is recommended that the state of Rhode Island implement a policy to manage Japanese knotweed along the Woonasaquatucket River.

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