Comparisons of Above- and Below-ground Carbon Storage in a Northeastern Illinois Urban Forest Following Rhamnus cathartica and Fraxinus spp. Removal

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Acknowledgements
I would like to thank Dr. Christie Klimas for her continued guidance and support throughout this project. I would like to acknowledge the financial support of an Undergraduate Summer Research Program (USRP) grant from DePaul University’s College of Science and Health. I would also like to thank Dr. James Montgomery, Dr. Liam Heneghan, and Stephanie Teresi for their help and direction.
Comparisons of Above- and Below-ground Carbon Storage in a Northeastern Illinois Urban Forest Following *Rhamnus cathartica* and *Fraxinus* spp. Removal

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**ABSTRACT** This study focused on quantifying potential differences in ecosystem services (carbon storage, soil organic matter, macroarthropod density) in a small, partially restored urban forest in order to determine if common buckthorn and standing dead ash removal effects can be detected while restoration is on-going. We calculated above-ground carbon storage (tons/total area) using whole tree biomass equations and compared this to i-Tree Canopy estimations. We collected SOM through loss-on-ignition and collected macroinvertebrates by pitfall trapping to determine differences along transects. Above-ground carbon storage, soil organic matter, and macroinvertebrate total results for this study were found to be statistically not significant, indicating that a short-term study is not an adequate measurement of these environmental indicators. Rather, these results reflect the varied findings of other short-term researchers and illustrate the importance of continued monitoring of ecosystem services following restoration, as results may be more accurate over a longer period of time.

**INTRODUCTION**

Invasive species in North America are disrupting ecosystem processes and devastating native species populations, increasing ecosystem instability (Boettcher, 2021). Restoration ecology is a growing scientific field that has developed in response to this anthropogenic damage, a large part of which focuses on invasive species remediation. From 1967-2017, the cost of invasive species remediation in North America was $1.26 trillion (Crystal-Ornelas, 2021), likely an underestimate.

Since there are currently 6,500 invasive species established in the United States as recorded by the U.S. Geological Survey (n.d.), about 23% of total species, more efficient methods of ecosystem management and observation are needed as both invasive species populations and the cost of their removal continue to expand. This is especially important in urban settings, where native ecosystems may be absent, degraded, or smaller in footprint,

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resulting in quick colonization and domination of invasive species in the few remaining green spaces. One such green space, urban forests, act as above- and below-ground carbon sinks, storing carbon in soil, roots, and biomass of living plants (Iannone III 2015; Mascaro & Schnitzer, 2011). Ecosystem services, like carbon sequestration or nutrient cycling, are valuable to dense, human communities (Klimas et al., 2016). Carbon sequestration is especially important as carbon dioxide and other emissions in urban environments can have a significant impact on human and environmental health, although more research is needed on this connection (Nowak & Crane, 2002). The restoration of urban forests can be a useful management tool to reduce rising atmospheric CO₂ levels as well as protect native and vulnerable species.

*Rhamnus cathartica,* or common buckthorn, is an invasive species introduced to the U.S. in the early 1800s as an ornamental shrub and recommended for windbreaks or hedges (WIGL 2021; Delanoy & Archibold, 2007). Due to the naturalization of this species into the North American landscape, it is now an aggressive invasive species that can quickly dominate an understory or take over prairie by forming dense monoculture thickets (Warren et al., 2017). Common buckthorn has been found to have a negative effect on native ecosystems through allelopathic inhibition of surrounding plants’ growth, speeding up decomposition of leaf litter, or decreased total forest biomass (Warren II et al., 2017, Heneghan et al., 2004). Furthermore, buckthorn presence can indirectly encourage the presence of other invasive species, such as earthworms (Heneghan et al., 2007). Buckthorn removal methods vary by region, but generally involve removal through physical or chemical means, and can also include replanting of native species (Delanoy & Archibold, 2007). These methods can be extremely costly or inefficient if not informed by seasonality, buckthorn density, and other factors (Delanoy & Archibold, 2007). Since buckthorn is extremely resistant to competition and restoration, it may have long-term effects that may be unexpected or hard to measure. Invasive species management is an ongoing process. To understand the effects of restoration, research quantifying and monitoring ecosystem services following restoration is important.

Indicators of forest productivity and health have been found to be impacted significantly by buckthorn, such as soil organic matter percentage (SOM) (Mascaro & Schnitzer, 2011). There is a positive correlation between larger trees and higher biomass and carbon storage, which also includes higher soil organic matter content (Mertes & Didier, 2021). In areas with a high density of buckthorn, total biomass and soil organic matter content are lower due to less native trees and more, smaller buckthorn shrubs (Hoff, 2015). Buckthorn also has a strong effect on nutrient cycling, speeding up decomposition rates and reducing vegetation species richness (Larkin et al., 2013). Furthermore, macroarthropod diversity is hypothesized to be impacted by buckthorn.

The research location in this study presents a unique site as this contiguous urban forest is currently divided into two sections based on the progress of restoration efforts in the area. Figure 1 shows Harbert Woods and the divide between the restored area where buckthorn was cut and covered (north-end) and unrestored area (south-end). The north section of the forest has been the focus of buckthorn topping as well as removal of standing dead ash (*Fraxinus* spp.) trees. Restoration efforts have not yet reached the south section which remains unrestored. These two areas provide an opportunity for researchers to monitor the long-term changes in ecosystem services and forest health following buckthorn removal and damage by the emerald ash borer (*Agrilus planipennis*). However, since this study
area is very small and the methods unreplicated across different urban forests, any observations can only be cautiously extended beyond this study.

Invasive species like buckthorn and emerald ash borers can have an indirect mutualistic relationship that can make the habitat less suitable for native species. By wiping out mature ash trees, borers leave open canopy for buckthorn to better establish and dominate a forest. It has been found that loss of ash species can result in patterns of negative human health effects, emphasizing the importance in restoring and protecting these native green spaces (Donovan et al., 2013).

**Figure 1.** I-Tree canopy generated map of Harbert Woods study area. The red line indicates the division between the unrestored southern area, which is heavily dominated by buckthorn, and the restored northern area.

**METHODS**

Harbert Woods is a 350m by 80m urban forest that is undergoing removal of buckthorn and standing dead ash trees followed by restoration. Around half of the site has been restored, with buckthorn being replaced with native plants (Figure 1). To compare above- and below-ground carbon storage by restoration status, two transects were placed in the unrestored section and two in the restored section. The transect lengths varied between 60 and 70m, with the restored section having smaller transect lengths than the unrestored section.

**Above-ground Carbon Storage**

In the four transects, we inventoried all trees (living and dead) above ten centimeters diameter at breast height (dbh). For each tree, we determined whether the emerald ash borer was the cause of death by the presence or absence of the distinctive paths taken by the beetle under the bark, leaving permanent indents in the deadwood. All inventoried trees were identified to species level. Based on allometric equations relating dbh to volume or aboveground tree biomass (Tritton & Hornbeck, 1982; Jenkins et al., 2004), we calculated the amount of carbon associated with each tree surveyed. Carbon storage was calculated similarly to the methods used in the “Carbon storage and valuation of ecosystem services on a restored urban forest in Northeastern Illinois” report (Mertes & Didier, 2021).

Carbon storage for the entirety of the unrestored and restored portions was found using the total tree biomass sum. This sum was converted from kilograms to short tons and multiplied by 0.5 to calculate the carbon equivalent. The carbon dioxide sequestration of the forest was found by multiplying the carbon equivalent by 3.67. The carbon dioxide product value was converted to metric tons using the multiplication of 0.90718474 and appropriately divided by the value of the percentage of Harbert Woods that was encompassed by each portion of the study area.

**Soil Organic Matter**

To quantify soil organic matter (SOM) percentage, 30 soil cores were taken along the four transects in the spring-summer of 2022, equidistant from each other along the transects. Prior to collection, leaf litter was removed from the sample area. For each core, the soil core sampler was lined with a plastic cylinder with a diameter of 3.8cm and length of 30cm and
hammered into the ground. Soil core sampling methods were similar to those described in Klimas et al. (2016). There were 30 total cores sampled.

To analyze the soil cores for organic matter, the loss-on-ignition technique was used (De Vos et al., 2005). The soil samples were air-dried, ground, and sieved before being prepared. They were then weighed into equal amounts in crucibles. The samples were placed in the muffle furnace at 500°C for 24 hours, then taken out when the furnace and samples were cooled. The samples were then weighed again. The post-ignition weight of soil and percent organic matter was then calculated using the following formulas.

\[
\text{post – ignition weight of soil} = (\text{post – ignition weight of crucible + soil}) – (\text{weight of crucible})
\]

\[
\% \text{ SOM} = \frac{\text{(pre – ignition weight of soil)} – (\text{post – ignition weight of soil})}{\text{pre – ignition weight of soil}} \times 100
\]

Macroinvertebrate Collection

Pitfall traps were set up to measure macroinvertebrate density by transect. Three traps were placed per transect, collected, and reset for three consecutive days. The traps were standard pitfall traps made from recycled yogurt cups (9.5cm diameter, 5.7cm depth), with a small amount of water and a drop of dish soap at the bottom. The traps were dug into the ground, flush with the surface of the soil. Once set, leaf litter and sticks were placed to cover the top partially and allow insects to travel over the trap (Figure 2). Since this study was done in a period with no rain, there was no rain cover necessary. The specimens collected were preserved in specimen jars containing 70% ethyl alcohol. They were then analyzed for total number of macroinvertebrates per transect.

Data Analysis

Data were exported from Microsoft Excel using a package by Bryan Wickham (2022). We tested soil organic matter and macroinvertebrate counts for normality using R. For both SOM and macroinvertebrate results, a fit distribution package by Delignette-Muller & Dutang (2015) was run to determine normalcy of the data. Then either ANOVAs or generalized linear models were run to determine whether there was a significant difference between restored and unrestored areas.

RESULTS

Above-ground Carbon Storage

The unrestored section of Harbert Woods had a calculated average of 171.88 (± 20.75) tons of carbon dioxide equivalent per acre (Table 1, next page) based on calculations using dbh and allometric equations. This value excluded dead ash trees. The restored section was found to have 375 tons of carbon dioxide per acre. The opposite result was found using i-Tree canopy software: results calculated 517±14.73 tons of carbon dioxide per acre in the unrestored section, compared to 218.4±10.66 tons carbon dioxide/acre in the restored section (i-Tree).

The mean of the biomass in the unrestored section was calculated to be 2738.17 kg of living biomass and 999.79 kg of standing dead. In the restored section, living biomass was found to be 10,868.93 kg and standing dead was 730.51 kg.
<table>
<thead>
<tr>
<th></th>
<th>Unrestored</th>
<th>Restored</th>
</tr>
</thead>
<tbody>
<tr>
<td>Carbon Storage Mean (T/yr)</td>
<td>171.88</td>
<td>375.69</td>
</tr>
<tr>
<td>STD</td>
<td>20.75</td>
<td>19.56</td>
</tr>
<tr>
<td>i-Tree Carbon Storage (T/yr)</td>
<td>517</td>
<td>218.4</td>
</tr>
<tr>
<td>SE</td>
<td>±14.73</td>
<td>±10.66</td>
</tr>
<tr>
<td>Biomass (kg)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Living</td>
<td>2738.17</td>
<td>10868.93</td>
</tr>
<tr>
<td>Standing Dead</td>
<td>999.79</td>
<td>730.51</td>
</tr>
<tr>
<td>Dead</td>
<td>10868.93</td>
<td>324.9</td>
</tr>
<tr>
<td>Mean</td>
<td>2099.0</td>
<td>53.18</td>
</tr>
</tbody>
</table>

Table 1. Aboveground carbon storage and biomass data.

Soil Organic Matter

Soil organic matter results were not statistically significant in their differences between treatments, having a higher average in the restored section of forest than the unrestored. The unrestored area had an average soil organic matter amount of 17.4%, while the restored area had an average of 21.3%. These results as well as the maximum and minimum SOM values are shown in Table 2.

<table>
<thead>
<tr>
<th></th>
<th>Average SOM (%)</th>
<th>Max. SOM (%)</th>
<th>Min. SOM (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unrestored 1</td>
<td>13.17</td>
<td>25.26</td>
<td>9.61</td>
</tr>
<tr>
<td>Unrestored 2</td>
<td>21.64</td>
<td>27.64</td>
<td>17.17</td>
</tr>
<tr>
<td>Restored 3</td>
<td>22.42</td>
<td>37.46</td>
<td>14.49</td>
</tr>
<tr>
<td>Restored 4</td>
<td>20.13</td>
<td>37.61</td>
<td>9.28</td>
</tr>
</tbody>
</table>

Table 2. Soil Organic Matter percentage by transect.

When analyzed in R, the percent soil organic matter results were normally distributed. However, analysis of variance results indicated there was no significant difference between treatments (Figure 3; p-value=0.159 Table 3).

Figure 3. Boxplots of soil organic matter percentage by restoration treatment. Maximum and minimum values are similar, but mean SOM is slightly greater for the restored treatment. The unrestored treatment has a greater interquartile range. The restored section has two outliers while the unrestored has none.

<table>
<thead>
<tr>
<th></th>
<th>Df</th>
<th>Sum Sq.</th>
<th>Mean Sq.</th>
<th>F value</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Treatment</td>
<td>1</td>
<td>107.9</td>
<td>107.9</td>
<td>2.091</td>
<td>0.159</td>
</tr>
<tr>
<td>Residuals</td>
<td>28</td>
<td>1444.7</td>
<td>51.6</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 3. ANOVA Results for Soil Organic Matter.

Macroinvertebrate Collection

Seventy-five total invertebrates were captured in the restored section and 32 in the unrestored section (Figure 4).
Fitdistr tests in R indicated that macroinvertebrate results were gamma distributed. Similar to the previous SOM results, the macroinvertebrate results were not significantly different (Figure 5; Table 4).

![Figure 4](image)

**Figure 4.** Bar graph depicting macroinvertebrate collection by day of collection. The most invertebrates were collected on day 1.

![Figure 5](image)

**Figure 5.** Boxplots of macroarthropod density by restoration treatment. The restored section features a greater maximum, minimum, mean, and interquartile range.

|          | Estimate | Std. Error | T value | Pr(>|t|) |
|----------|----------|------------|---------|---------|
| Intercept| 0.04     | 0.019      | 2.126   | 0.101   |
| Treatment Unrestored | 0.054 | 0.048 | 1.121 | 0.325 |

**Table 4.** Generalized linear model results for macroinvertebrate data.

**DISCUSSION**

In this study, there were no significant differences in above-ground carbon storage, SOM, and macroinvertebrate density. Other researchers have found similarly modest results, which reflect the inability for recently restored ecosystems to change factors like carbon storage or insect diversity quickly. For example, a similar short-term study on *R. cathartica* impact on invertebrate diversity found no significant differences in abundance or species richness (Chartier et al., 2020). However, other researchers who have studied restored woodlands decades after restoration have found significant differences in leaf litter mass, but no significance in soil carbon storage measurements (Zirbel et al., 2011). This expresses that ecosystem indicators like carbon storage change slowly over time after restoration, requiring long-term studies to adequately address impact.

Above-ground carbon storage has been found to be reduced by buckthorn colonization and replacement of native trees and understory plants (Mascaro & Schnitzer, 2011). Buckthorn’s limited maximum size compared to native mature trees results in limited capacity for carbon storage within biomass and a statistically significant higher overall biomass in native canopies (Mascaro & Schnitzer, 2011). An important aspect of invasive species restoration is understanding species traits that allow invasive species to become so prevalent, such as taking advantages of available gaps in forests or discouraging surrounding plant growth through root cytotoxins (Warren II et al., 2017). The unrestored section of Harbert Woods displays the success of invasive species characteristics, as it is a densely packed buckthorn stand with few mature native trees. However, any measurements, such as i-Tree carbon estimates, (although not significant) of higher above-ground carbon storage and biomass in the restored section of Harbert Woods is most likely due to a higher density and total count of mature, living trees in the restored section. In the future, more wide-reaching and collaborative studies within different urban forests may result in stronger results.

In a recent short-term study focusing on a forest that had undergone restoration since the 1980s, Larkin et al. (2013) found SOM increased over the years following repeated restoration from buckthorn-invasion as replanted native...
understory species were able to better contribute to SOM cycling. However, the results expressed had high unexplained variance. Since results are difficult to quantify until possibly years after initial restoration, it is important that researchers revisit restored or partially restored green spaces. As Larkin et al. (2013 p. 7) states, “Observed ecosystem changes came after what were not just one-time restoration actions but restoration followed by years of on-going, active management.” Especially regarding invasive species-affected and unstable urban ecosystems, time is needed to adequately assess ecosystem services or quality of restored sites. Harbert Woods may be showing very preliminary signs of improvement, but there is much work to be done to achieve definitive or significant results.

Past researchers have found varying effects of buckthorn on soil properties like nutrient cycling, decomposition, SOM, and micro/macroarthropod diversity, underscoring the diversity of factors that invasive species affect. Heneghan et al. (2002) found that leaf litter decomposition rates were significantly different between treatments, which may impact the diversity and density of microarthropods in the area. Buckthorn was found to attract some insects earlier than native trees, but leaf litter was also decomposed earlier, which can be detrimental to remaining arthropod populations (Heneghan et al., 2002). These results were strengthened by Iannone III et al. (2015), who found that buckthorn establish in places with greater spring leaf litter mass and greater rates of decomposition and subsequently further accelerate decomposition. These changes to soil ecosystems can be long-lasting, even during and after restoration, complicating the ability short-term studies to capture improvement in ecosystem health.

Buckthorn has been found to have an indirectly mutualistic relationship with other invasive species, such as European earthworms or emerald ash borers. In a study by Roth et al. (2015), buckthorn and earthworms have been found to facilitate each other’s invasion, resulting in co-invasion. The researchers compared differences in buckthorn germination in manipulated soil, leaf litter, and earthworm density conditions and determined that earthworm presence was the most important factor in germination (Roth et al., 2015). This behavior of co-invasion could help guide future restoration efforts. This secondary facilitation between invasive species is also present between emerald ash borers and buckthorn (Baron & Rubin, 2020). As buckthorn occurrence was found to be significantly higher in canopy gap areas, the death of mature trees by emerald ash borers may provide the space for buckthorn invasion in gaps that may have not been present otherwise (Baron & Rubin, 2020). Although not the focus of this study, removal of standing dead ash may have a positive effect on buckthorn spread if not addressed and should be monitored after removal efforts (Baron & Rubin, 2020).

This study represents an important collaboration between researchers and the local community, as community restoration efforts allowed for research based upon restoration status comparison. There are still gaps in understanding the relationship between invasive species, restoration, and human health consequences that can be addressed collaboratively (Heneghan et al., 2008). Due to the complexity and longevity of ecosystem dynamics, restoration methods and results must be continuously studied and improved. As Heneghan et al. (2004) states, soil property or biodiversity changes may result in restoration methods having to adapt to new situations and resilience. It is essential that future research is informed by ecosystem service indicators and monitors invasive species impact, as well as involving local communities to ensure long-term effects of restoration on the environment.
ACKNOWLEDGEMENTS

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Sanchez-Dudik and Drosos: Comparison of carbon storage following Rhamnus cathartica removal


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