

2016

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Sam L. Pepper

*DePaul University*, [spepper37@gmail.com](mailto:spepper37@gmail.com)

Liam Heneghan

*DePaul University*, [lhenegha@depaul.edu](mailto:lhenegha@depaul.edu)

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### Recommended Citation

Pepper, Sam L. and Heneghan, Liam (2016) "Implications of Manipulations of Soil Quality on the Growth of European Buckthorn (*Rhamnus cathartica*) in a Greenhouse," *DePaul Discoveries*: Vol. 5 : Iss. 1 , Article 14.

Available at: <https://via.library.depaul.edu/depaul-disc/vol5/iss1/14>

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## Implications of Manipulations of Soil Quality on the Growth of European Buckthorn (*Rhamnus cathartica*) in a Greenhouse

Sam Pepper\*

Department of Environmental Science and Studies

Liam Heneghan, PhD

Department of Environmental Science and Studies

**ABSTRACT** European buckthorn (*Rhamnus cathartica* L.) is an invasive shrub that is capable of changing the nitrogen content of a soil through the deposition of its nitrogen rich leaf litter. This change creates conditions that may favor recolonizing by buckthorn and negatively affect the growth of native plants. In this greenhouse experiment, we examined the effect of altering soil quality by adding mulch comprised of buckthorn wood on above and belowground biomass, stem length and leaf production of buckthorn saplings. We found that buckthorn saplings planted in buckthorn mulch had reduced stem length and leaf production compared to growth in control sites. Extrapolating from these results we suggest that amending soils in sites set aside for biodiversity conservation and restoration may result in reduced growth of this invasive shrub.

### INTRODUCTION

Invasive species have the potential to significantly impact unique ecosystems by threatening the native species and by modifying ecosystem services that provide economic revenue for the region (Becker et al. 2013). Although these ecosystems are threatened by a range of anthropogenic impacts including altered hydrology, loss by fire, and so on, invasive species are problematic because they are capable of altering plant and animal assemblage structures and the a range of key soil ecosystem

processes in the habitats that they invade (Heneghan et al. 2004).

European buckthorn (*Rhamnus cathartica* L.) has received attention from researchers and restoration managers in the US Midwest because it is a prevalent woody shrub that can outcompete native species for resources. European buckthorn became popular in North America in the late 19<sup>th</sup> century as a decorative shrub for fences and hedges (Kurylo et al. 2012). Profession societies that promoted agriculture in the region at this time period praised buckthorn's superiority and hardiness over other fence and hedge species (Kurylo et al. 2012).

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\* Corresponding Author: [spepper37@gmail.com](mailto:spepper37@gmail.com)  
Research Completed in Fall 2015

Although it was introduced into the region more than one hundred years ago, it did not become prevalent in sites set aside for conservation until the 1970s.

A review by Knight et al. (2007) suggests that European buckthorn has unique physiological characteristics which enables the shrub to outcompete native species. These characteristics include rapid growth, high photosynthetic rates, wide tolerance ranges for moisture and drought, and unusual phenology which may provide it with an advantage over native species. This gives it an advantage over native species within the invaded environments and thus sites dominated by European buckthorn have consistently fewer native species. Knight et al (2007) acknowledge the difficulties in quantifying the effects of European buckthorn on native species since the factors they examined do not appear to fully explain the magnitude to which buckthorn affects native plants and animals. There is, they claim, still much to learn about the impacts of buckthorn on above and belowground systems.

European buckthorn produces leaf litter high in nitrogen which decomposes rapidly compared with many native species. As a consequence, soils associated with buckthorn may have higher available nitrogen (Heneghan et al 2006). In addition buckthorn may modify soils in other ways. For example, a greenhouse experiment by Klinosky et al. (2013) compared the effects of European buckthorn foliage and *Acer saccharum*, a native tree to southern WI that has a similarly high nitrogen content in its foliage. They concluded that buckthorn inhibited germination of other plant species, and showed that litter produced by buckthorn inhibits the growth of the seedlings of four native trees (Klionsky et al. 2013). Thus buckthorn may regulate invaded systems by modifying soil conditions.

Since buckthorn modifies the soils in which it grows, there has been interest in examining ways to amend the soil as a means of reducing the growth of buckthorn and promoting the health of native plant communities. For example, in a 3 year field study undertaken by Iannone et al. (2013) they assessed the

implications of applications of buckthorn mulch on its own reinvasion potential. The study found that, contrary to expectations, that tilling European buckthorn mulch into the soil increased the nitrogen availability in the soil (Iannone et al. 2013). However, their results showed that there was an 82% reduction of biomass when European buckthorn mulch was tilled into the soil than when it was laid across the surface (Iannone et al. 2013). They recommended that future research of this soil amendment should isolate the effect of mulch from tilling. Thus, our research looks at the implications of mulch additions for buckthorn growth in the greenhouse. The study is designed to look at mulch without the confounding effect of physically tilling the buckthorn into the soil.

In our study, we examined how manipulating soil quality using European buckthorn mulch can affect the growth of European buckthorn in a greenhouse setting. We hypothesized that application of European buckthorn mulch in the soil would reduce the growth of European buckthorn saplings. Although we did not directly measure this, we speculated that the mechanism determining the reduced growth was that the mulch which has a high carbon to nitrogen ratio would reduce the nitrogen available to the growing plants.

## METHODS

European buckthorn saplings were collected at McDonald's woods, at the Chicago Botanic Garden, Glencoe, Illinois. A total of 29 saplings were transported from the site with intact root systems. The root systems were cleaned as much as was feasible of soil from the site of origin. No pruning was conducted on the saplings once they were placed in their planter pots.

These saplings were placed in three different soil treatments. The first treatment was used as the control and comprised of potting soil (100% MetroMix = unmulched); the experimental group consisted of 2 treatments of European buckthorn mulch (50% European buckthorn mulch and 50% Metromix = half mulch and 100% European buckthorn mulch = full mulch). The experimental group of 50% European buckthorn mulch was homogeneously mixed in the planter pots. The European buckthorn mulch

was prepared from cut European buckthorn shrubs that were collected from a restoration project at the River Trail Nature Center (3120 N. Milwaukee Avenue, Northbrook, Illinois). The collected European buckthorn branches were ground-up in a wood chipper. Woodchips that were mixed with and without potting soil had a high porosity. The volume of soil within each planter pot is approximately 211 cm<sup>3</sup>.

The experiment was established in the DePaul University greenhouse on May 8th 2015 and continued until September 27th 2015. Samples were randomly assigned a position on a 248 X 126 cm table. Measurements of stem length and leaf production were taken from each individual plant biweekly over the course of the experiment. The stem length was measured between the base of the stem to the terminal bud of the samples with a 30 x 30 SI fiberglass measuring tape. Leaf production was measured by counting the individual leaves produced by the sapling. We were interested in the total number of leaves on the plant on a bi-weekly basis.

At the end of the experiment aboveground and belowground mass was also measured. We measured the aboveground and belowground masses because we are interested in the overall effect of the treatments on European buckthorn growth. The belowground mass measured the root system of the saplings. The root systems were prepared by carefully hosing off the samples of the soil attached to the root system. A No. 70 sieve was used to capture the soil from clogging the drains. In order to weigh the root

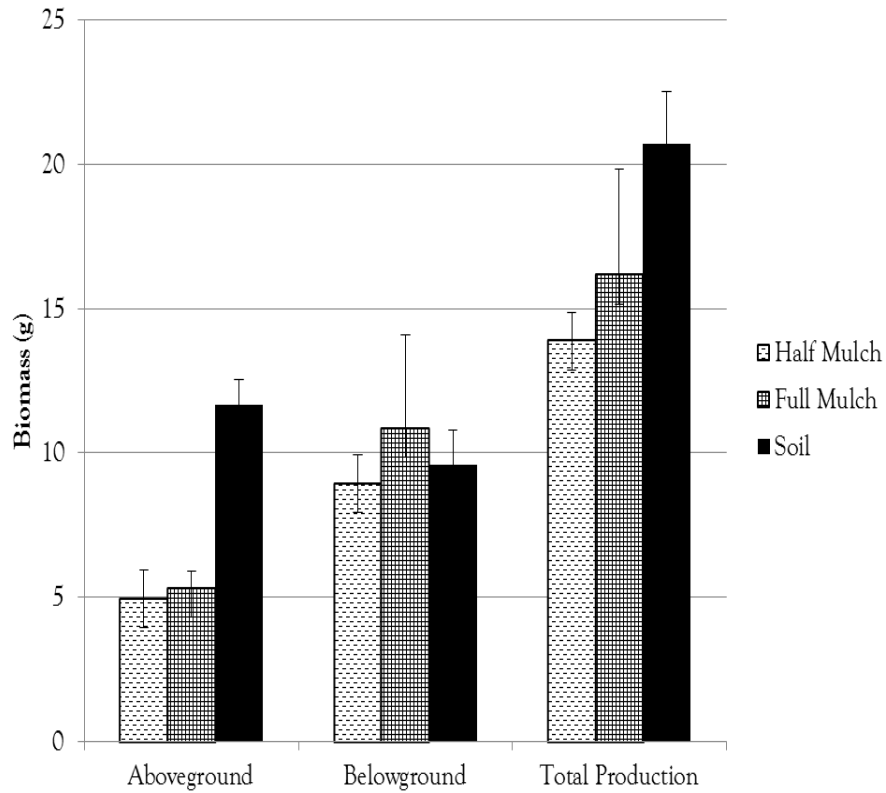
system without completely damaging the sample the wood chips remained in the root system. We shall devise a more thorough way of isolating roots for future work. Aboveground mass was measured by separating the stems from the root systems and clipping the stems into segments that could fit into a plant potter. Aboveground and belowground measurements required a plant potter that was tiered and weighed on a PB 403-S Mettler Toledo scale.

Stem length and leaf production between European buckthorn mulch treatments and the control were in each case analyzed using ANOVA.

## RESULTS

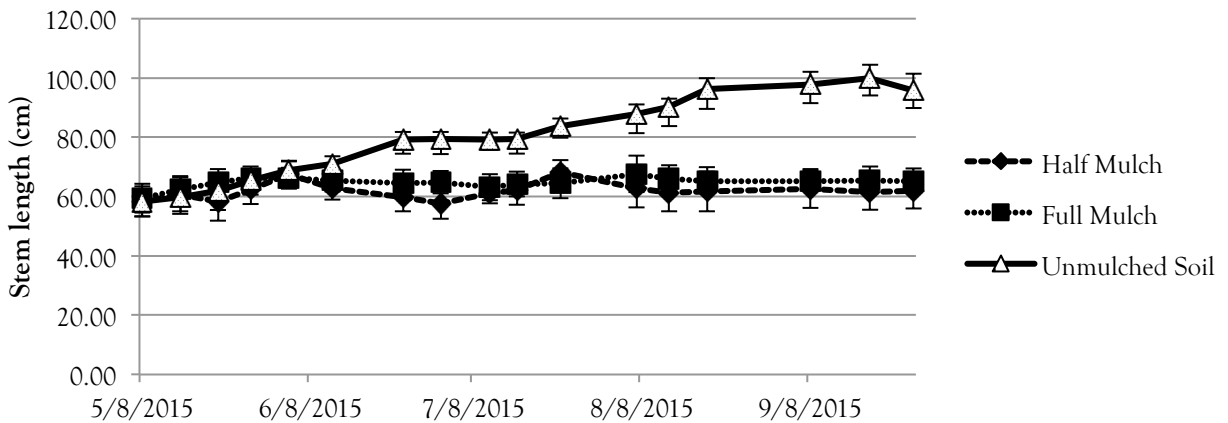
The aboveground biomass was highest in control soils and lowest in the half and full European buckthorn mulch plots (Figure 1). There was not an overall difference in the belowground biomass between unmulched, half, and full mulched plots. The total biomass of the plots was higher in soil compared to buckthorn mulch treatments. Stem length (Figure 2) and leaf count (Figure 3) were highest in plots planted in unmulched soil, and lowest in plots planted with half and full European buckthorn mulch. There was a difference in stem length between the unmulched samples and the half and full European buckthorn mulch samples (Figure 2). The ANOVA analysis for leaf production reported  $F= 5.05$  and  $p = 0.0144$ . The ANOVA analysis for stem length reported  $F= 8.86$  and  $p<0.0013$ .

## Final Biomass

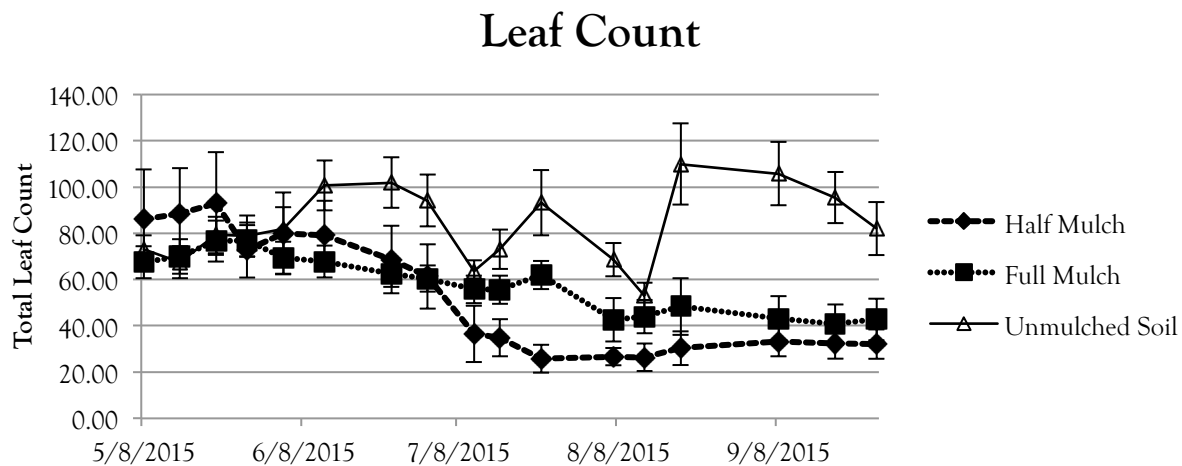


**Figure 1:** The aboveground, belowground, and total biomass (g) at the end of project. Bar graph portrays average weights.

## Stem Length (cm)



**Figure 2:** Stem length (cm) of half mulch, full mulch, and unmulched saplings throughout the duration of the experiment. Each point represents the average stem length on the day of measurement.



**Figure 3:** Leaf count of half mulch, full mulch, and unmulched saplings taken throughout the duration of the experiment. Each point represents the average leaf count taken on the day of measurement.

## DISCUSSION

This experiment showed that the application of European buckthorn mulch can reduce the aboveground biomass, stem length, and total leaf production of European buckthorn saplings.

The results reported here also show that there is no difference between treatments constituted completely by mulch or half mulch and half soil. The application of European buckthorn mulch in both treatments may have altered the available soil nutrients by reducing the available nitrogen for the plant (Iannone et al. 2013, Iannone et al. 2015, Klinosky et al. 2013, Knight et al. 2007). Thus by adding the mulch the fertility of the soil is reduced and diminishes the saplings capacity for growth.

Although this conjecture that the results obtained in the experiment emerged from a lowering of soil quality that accompanies the addition of mulch, this remains to be confirmed. We will investigate this by conducting a chemical analysis on the soils which we have preserved from the experiment.

Although we speculate that diminished growth of buckthorn saplings resulted from a lowered availability of nitrogen in the soil, there are

alternative mechanisms that might have been important. Plants rely on chemical signals to perceive the environment around them and send these signals to the above and belowground systems (Spence et al. 2015). Abscisic acid is biosynthesized directly in the shoot system of a plant and plays an important role for plants to respond to abiotic and biotic stresses (Spence et al. 2015). Planting European buckthorn in mulch composed of itself could have stimulated an acceleration of abscissions, causing the leaves to shed quicker (Spence et al. 2015). This would result in lower sapling productivity

The results we found in this study could have implications for restoration management in the field. In the past applications of European buckthorn mulch have been implemented through tilling. Iannone et al. (2013) suggests that tilling itself could be the most effective measure to reduce the growth of European buckthorn. Tilling physically alters soil ecological processes. Though tilling could reduce invasive species in areas set aside for nature, tilling could degrade native populations within the area by accidentally disturbing undetected seed banks (Iannone et al. 2013).

Unnecessary damages to native species could be a drawback to restoration management projects (Gassman et al. 2013). Even though these are plausible consequences, mulch additions without tilling may be more effective, and this possibility needs to be investigated.

Mulch amendments can potentially provide restoration managers with tools that are an economically viable alternative to more laborious restoration strategies. The laborious strategies include repeated revisits to a site to remove generations of reinvading shrubs. If managers chose to add European buckthorn mulch in a site it will be important to closely monitor the outcomes of such field manipulations.

In order for these results we present here to provide insights into novel methods for controlling the spread of European buckthorn, it will be necessary to show that European

buckthorn mulch slows growth for this exotic shrub more than it does for completing native shrubs. Native shrubs tend to be more adapted to low nitrogen conditions than invasive shrubs. This follow up work will be conducted starting in spring 2016.

## CONCLUSION

Our experiment found that the application of half or full European buckthorn mulch within a soil composition reduced aboveground biomass, stem length and leaf production of European buckthorn saplings. In order to fully understand the mechanisms determining the reduced productivity we must conduct a chemical analysis on the soil. More research is required to determine what affects European buckthorn mulch would have in a restoration site and native species. Research on these issues will be conducted in spring 2016.

## ACKNOWLEDGEMENTS

This work was funded by DePaul University. This project would not have been possible without the help of Margaret Workman, the employees of the River Trail Nature Center for allowing us to collect their cut buckthorn, Hakim Akhtar for assisting with the mulching, and Saif Al Muhairi and Rosie Fitz for helping to water the samples.

## AUTHOR CONTRIBUTIONS

S.P. created experiment proposal, recorded data, and tended the saplings and L.H. contributed to data analysis results.

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