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Joerger, Jessica; Logan, Tanner; Kious, Kiefer; and Rancic, Jake. "The Effects of Simulated Herbivory on the Growth of Wisconsin Fast Plants" (2016). *Celebration of Learning*.

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L1-G5 - Jake Rancic, Kiefer Kiou, Jessica Joerger, & Tanner Logan

Effect of simulated herbivory on the growth of Wisconsin Fast Plants

Abstract:

Wisconsin fast plant (*Brassica rapa*, *Brassicaceae*) seeds were planted in three six-celled containers with 15 seeds in each tray. After a week of germination in standard potting soil and tap water, we started our manipulation of simulating herbivory with scissors. E1 plants had both cotyledons of each plant removed, E2 plants had half of its cotyledons each plant removed, and the control group was allowed to grow. E2 plants had its foliage leaves cut when they exceeded 1 cm from that week on. Every seven days, the height of all the plants were recorded as well as the number of flowers and foliage leaves. By the fifth week, the average height (mm) of the control plants was 145.8, E1 plants was 100.8, and E2 plants was 96.3. The average number of flowering plants during the fifth week of measurement was 9 plants for the control group, 9 for E1, and 6 for E2. The t-test for the height of control plants vs E1 plants was statistically significant ($p < 0.018$). The control plants vs E2 plants was also significant ($p < 0.0017$). The E1 vs E2 plants was not significant ($p < 0.755$). After five weeks the plants were clipped and weighed. E1 had a final weight of 0.022g, E2 0.016g, and the control 0.050g. The manipulations and the control had significant p-values ($p < 0.0009$ for both) but the comparison between E1 and E2 was insignificant. Our hypothesis that plants which experience a loss of half of their cotyledons (E1) will have a significantly lower biomass than the plants that undergo continuous herbivory (E2) was partially supported through our data. We suspect that herbivory, regardless of whether it is continuous, may have an equal effect on the fast plants because we failed to reject the null hypothesis.

Introduction:

The Wisconsin fast plant is a herbaceous perennial with a generally short life cycle. In previous studies it has been found to flower in 2 to 3 weeks after adequate planting and has a life cycle of roughly 40 to 50 days. (Goldman 1999). One of the main concerns in plant growth is herbivory. Cotyledons are formed along with the root and shoot meristems, and are therefore present in the seed prior to germination. In general they are found to provide nutrients and energy to the developing seedling (Sotelo et. al. 2014). The effects of herbivory on cotyledons and the subsequent growth of mature plants has been demonstrated by (Hanley and Fegan 2007) who examined the effect of severing the cotyledons mechanically with scissors to replicate herbivory of seven dicotyledonous grassland species at intervals of 7, 14, and 21 days. They maintained a control from each species which did not receive treatment. The results of this study indicate that cotyledon removal greatly reduced growth for all species that were tested and that earlier cotyledon removal corresponded to greatest long-term effect on plant performance (Hanley and Fegan 2007). Plants in the experimental group with the cotyledons severed after seven days consistently had lower growth and a lower biomass turnout than the control. Regardless of species, the biomass of the 7 day experimental plants at 35 days old was less than half of the results of their control group. This study indicated that cotyledon removal early in a plant's life has detrimental effects on its growth (Hanley and Fegan 2007). In another study, Karban and Strauss observed the effects of herbivores on growth and reproduction of perennial host plants. Thrips, moths, and spittlebugs acted as the herbaceous organisms in the study. Herbivory affected aboveground and belowground plant growth and root:shoot ratio saw a decline. Vegetative growth did still occur, but flower head production did see a decrease overall (Karbon and Strauss 1993).

In an additional study by Lowenberg, researchers examined the effects that herbivory from deer and pollen feeding mites had on pollination in relation to flower growth. It was observed that flowers affected by herbivory had decreased ovule counts in their flowers lowering the chance of fruit-set. In fact some of the flowers *Pastinaca* and *Heracleum* changed gender and growth of sexual organs after herbivory. Overall plant biomass and growth was negatively affected after herbivory which also decreased appearance and size of flowers.

Based on the desirable characteristic traits of the fast plants, our goal will be to see how simulated herbivory affects the growth of the plant over its relatively short life cycle. Results of other studies with similar variables and procedures leads to our hypothesis. The plants that experience a loss of all of their cotyledons will have a lower biomass than the plants that undergo continuous herbivory.

Methods:

We obtained three planting trays and filled each with an adequate amount of soil. The soil was compacted 2 mm short of the rim of the trays. Each tray had alternating numbers of two and three holes in each cell, in which to plant the Wisconsin fast plant (*Brassica rapa*) seeds. The holes were approximately 2-3 mm deep from the surface of the soil. In each hole one seed was planted, with a total of 15 seeds per tray. Soil was placed over the seeds and the trays were then placed under the growing lamps. We added enough water to the tub to bring the water level to three centimeters. The plants were given a week to grow before beginning experimentation.

One potting tray was marked as the control group and the remaining two trays were marked as Experiment 1 (E1) and Experiment 2 (E2), respectively. After one week each plant's height was individually recorded along with other physical observations (discolorations, inforamities). Both cotyledons of E1 were removed, while in E2 only half of each of the

cotyledons and half of the foliage leaves were removed. The plants were then watered regularly and height measured every Tuesday. Following the five week observational period the plants were clipped at soil level and weighed individually. Mean values were calculated and obtained for each group before further analysis was started. A student T-test was performed to determine significance.

Results:

The initial height measurements for the fast plants averaged 31.82 mm for E1, 27.72 mm for E2, and 33.79 mm for the control. Only the comparison between E2 and the control for the initial height measurements was statistically significant ($p < .05$). After five weeks the final heights averaged 100.8 mm for E1, 96.357 mm for E2, and 145.8 mm for control. The differences between the final heights of the control was statistically significant when compared to E1 ($p < 0.018449$) and E2 ($p < 0.001709$). The difference between E1 and E2 were not statistically significant with $p < 0.7556$ (Figure 1). Comparisons between final E1:Control and E2:Control were both statistically significant ($p < 0.018449$ and $p < 0.001709$ respectively), however comparisons between E1 and E2 were not (Figure 2). The differences in height between plants at Week 1 and Week 5 were very statistically significant across all treatments: E1₁:E1₂, E2₁:E2₂, Control₁:Control₂ (Table 1).

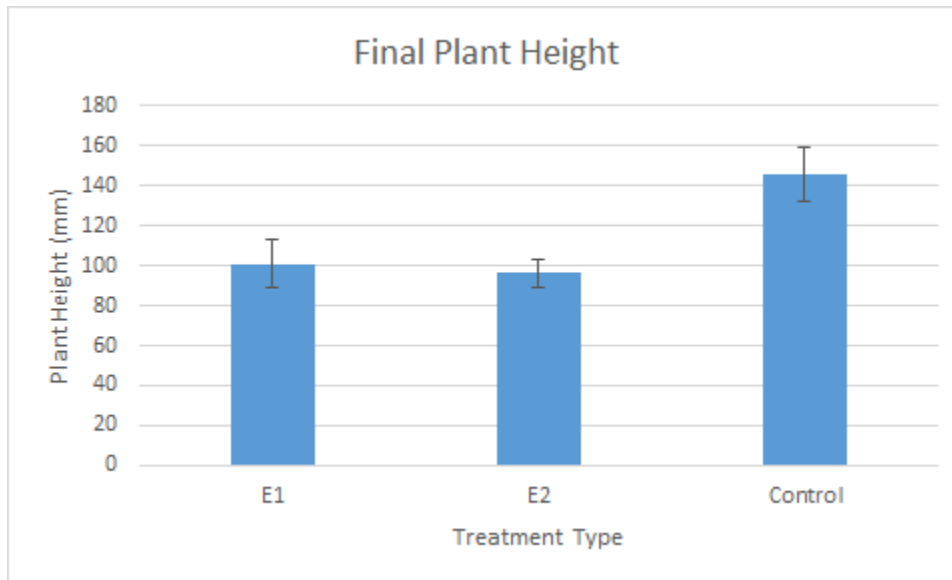


Fig. 1. Plant height at the end of five weeks of E1, E2, and Control. P-values between E1 and E2 were not statistically significant while the p-values between the treatments and the control were.

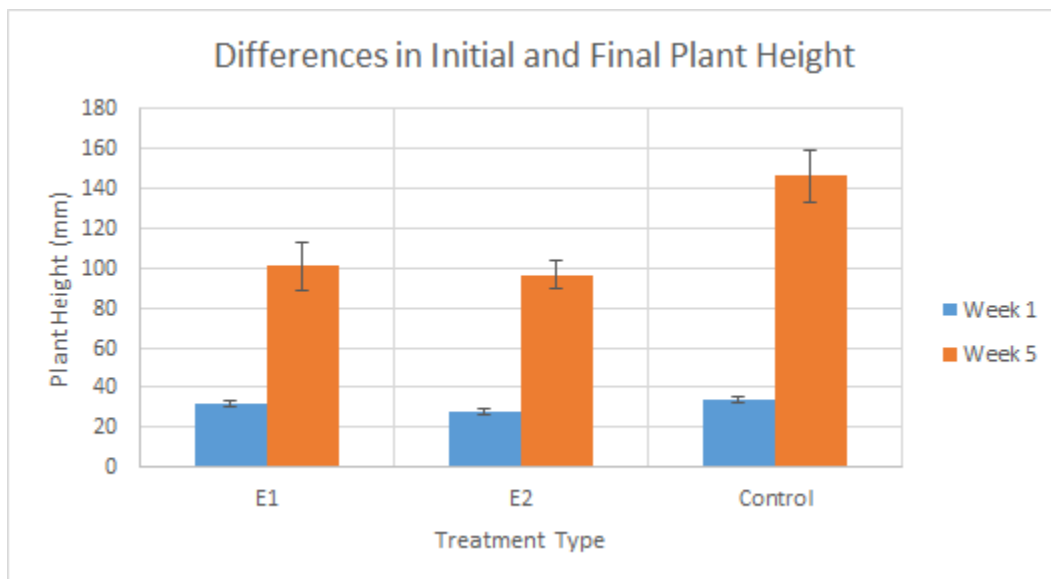


Fig. 2. Differences in final (Week 5) versus initial (Week 1) plant height measurements. In both Week 1 and Week 5 the differences between the control and treatment groups were statistically significant while the differences between the experimental treatment groups were not.

P-values for initial E1 and E2 were statistically insignificant at $p < 0.106$ as were the values for E1 versus control at $p < 0.437$. The comparison between E2 and the control was statistically significant with a p-value of $p < 0.022$. The differences between the final heights of the control was statistically significant when compared to E1 ($p < 0.018449$) and E2

($p < 0.001709$). The difference between E1 and E2 for Week Five was not statistically significant: $p < 0.7556$.

After five weeks of simulating herbivory, the final weights for the fast plants averaged 0.225 g for E1, 0.23 g for E2, and 0.4623 g for Control (Figure 3). Comparisons between E1:Control and E2:Control were both statistically significant ($p < 0.0009$ for both) but E1:E2 was not (Figure 3).

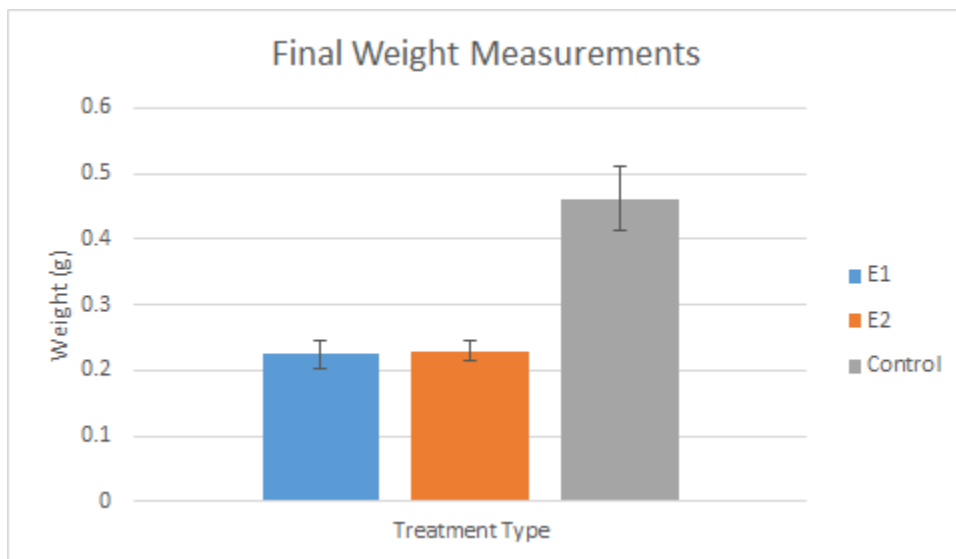


Fig. 3. Week five average weight calculations. The comparisons between all three treatments were found statistically significant E1:Control ($p < 0.000926$), E2:Control ($p < 0.000891$), and E1:E2 ($p < 0.856$).

T-test Initial versus Final Values		
E1w1:E1w5	E2w1:E2w5	Controlw1:Controlw5
4.31507E-05	4.88474E-08	1.613E-06

Table 1. T-test of final versus initial height values based on treatment type. W1 and W5 represent Week 1 and Week 5 respectively. All values are significantly lower than the statistical requirement of $p < 0.05$ and so the differences between the initial and final growth are all statistically significant.

During weeks 4 and 5 flower development was observed in all three of our test groups.

Over these two weeks we recorded the number of flowers visibly seen from plant to plant.

During week 4 collection we counted E1 as having 4 visible flowers, E2 as having 9, and the Control group as having 8. During week 5 the E1 group had a higher flower count of 9, E2 had a lower flower count of 6, and the Control group had a higher flower count of 9 flowers (Figure 4).

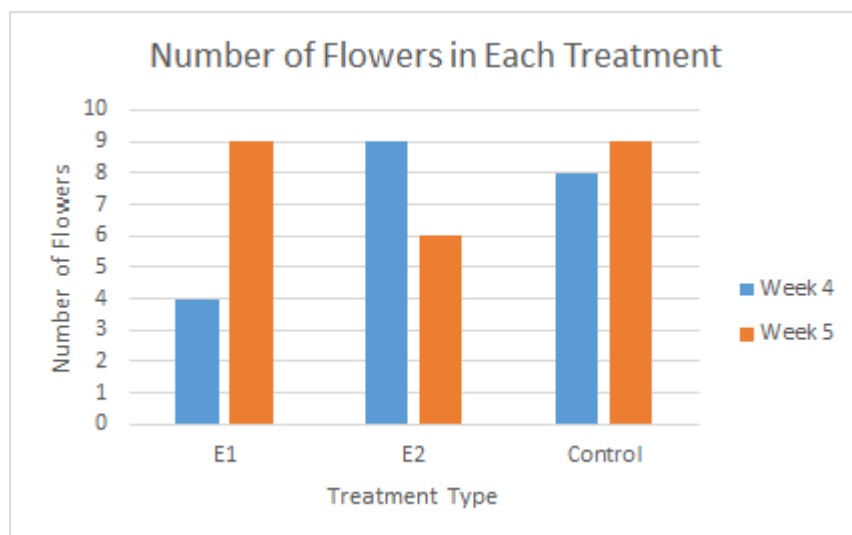


Figure 4. Comparison of the number of flowers by treatment type from the fourth and fifth week of measurement. Results indicate a growth in flower number for the control and E1 groups between week four and week 5 while a decrease in flower number for E2.

Discussion:

Upon examination of several studies in relation to herbivory effects on plant growth, our experiment performed on the Wisconsin Fast plants produced similar results. Hanley and Fegan's (2016) findings are consistent with our results because the fast plants that experienced simulated herbivory had lower average heights after five weeks than the control. Similarly Karban and Strauss' study agrees well with our results in that our E2 flowers that experienced weekly herbivory showed a decrease in flower production. This result also corroborates Lowenberg's (1997) study because the manipulated groups (E1 & E2) had a lower overall biomass than the control group. In Sotelo et. al. (2016) study on plant responses to mild herbivory, it was shown that the *Brassica rapa* plants had increases in bulb mass and

overcompensated their leaf growth to combat the effects of herbivory. In our study in group E1 we observed that the plants increased leaf production and foliage size after simulating herbivory. Overall, after examining the results of our own study performing simulated herbivory on *Brassica rapa* and several other studies with similar conditions, we conclude that simulated herbivory has an overall negative effect on plant growth in Wisconsin fast plants.

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