# A Review of Previous and Ongoing Studies of Intercropping as an Alternative to Fertilizers and Pesticides

University of Michigan Biological Station

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6/10/13

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Intercropping is a technique studied by agroecologists in which two or more species are planted together to improve agricultural systems. Since industrial agriculture has proven to be ecologically destructive, the reintroduction of intercropping in United States farming is part of the larger effort to create a sustainable and secure agricultural system. Intercropping improves the use of resources including water, soil, and labor. This paper focuses on intercropping as an alternative to pesticides and nitrogen fertilizers. In addition to reviewing other studies, we analyzed our own study in which we intercropped arugula with beans and marigolds. We hypothesized that arugula yield would be greatest when intercropped with beans and marigolds since beans increase nitrogen availability and marigolds deter herbivory. Though the results suggested that beans inhibit the growth of arugula in comparison to the control, all treatments with marigolds had significantly less herbivory.

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# A Review of Previous and Ongoing Studies of Intercropping as an Alternative to Fertilizers and Pesticides

University of Michigan Biological Station

**EEB 381: Introduction to ecology** Joel Heinen

6/10/13 Kapustka, Kiseleva, Tryban, Yates

#### **Abstract**

Intercropping is a technique studied by agroecologists in which two or more species are planted together to improve agricultural systems. Since industrial agriculture has proven to be ecologically destructive, the reintroduction of intercropping in United States farming is part of the larger effort to create a sustainable and secure agricultural system. Intercropping improves the use of resources including water, soil, and labor. This paper focuses on intercropping as an alternative to pesticides and nitrogen fertilizers. In addition to reviewing other studies, we analyzed our own study in which we intercropped arugula with beans and marigolds. We hypothesized that arugula yield would be greatest when intercropped with beans and marigolds since beans increase nitrogen availability and marigolds deter herbivory. Though the results suggested that beans inhibit the growth of arugula in comparison to the control, all treatments with marigolds had significantly less herbivory.

#### Introduction

Agroecology is the study and application of ecological principles to agricultural systems in order to improve yields and resource use in an environmentally sound way (Gliessman, 1990). The importance of this field grows as the human population increases and requires a more efficient and sustainable food supply. One topic agroecologists study is intercropping, a farming technique in which two or more species are planted together to improve growing conditions. This practice has its roots in Southeast Asia and Latin America, such as the planting of corn and cassava in the Philippines (Vandermeer, 1992). The combination improves land use, as corn grows quickly and can be harvested before the cassava is mature (Ayoola, 2011). Scientific research of intercropping in United States began in the 1910's when studies showed the benefits of intercropping in grape vineyards and lettuce fields (Husmann et. al., 1916; Watts, 1917). Other studies showed the negative effects of intercropping which stem from competitive interactions between intercropped plants (Hauggaard-Nielsen, 2004; Cenpukdee, 1992). Longterm research shows that intercropping has fewer negative implications compared to today's industrialized agriculture that focuses on monocultures (Nelson, 1998). We investigated intercropping arugula, beans and marigolds in the garden of the University of Michigan Biological Station (UMBS). We had two hypotheses: first, that leaf area would be greater in Treatments 2, 3 and 4 than in the control, Treatment 1; and, second, that herbivory levels would be lower in the treatments with marigolds (Treatments 3 and 4) than in the treatments without marigolds (Treatments 1 and 2). The continued research of intercropping and agroecology in general is necessary to further the understanding of plant interactions, improve agricultural ecology and strengthen food security.

# **Development of Intercropping within Agriculture**

Intercropping was practiced in traditional societies for centuries. Early evidence of polyculture comes from Theophrastus' book *On the Causes of Plants*. He discusses the benefits of planting barley or wheat together with olives or grapes (Lithourgidis, 2011). The Southwestern indigenous American Tribes used intercropping to grow three of their major food crops: beans (*Phaseolus vulgaris*), maize (*Zea mays*) and squash (*Cucurbita pepo*) (Harty, 2008). The low growing squash acts as a ground cover to protect the other crops from moisture loss while the tall stalks of corn support the growing of beans. The beans, of the Fabaceae family, fix nitrogen in the soil, improving the growing conditions for other plants. Other examples of intercropping around the world include planting soybeans with rice in India, peas with oats in Germany and rice with sugar beets in China (Vandermeer, 1992).

Intercropping was also common in the United States until the invention of industrial agriculture after World War II (Horwith, 1985). The monocultures in industrial agriculture lack the benefits of polycultures and thus require the use of pesticides and fertilizers. The term agroecology began to make its way into literature during the 1970's, defined as the "global study of agroecosystems protecting natural resources" (Wezel, 2009). Viewing agriculture as its own ecosystem, agroecology aims for sustainable yet optimal approaches to farming, including alternative agriculture strategies such as intercropping (Gliessman, 1998).

# **Problems with Current Industrial Agriculture**

Industrial agriculture severely degrades many ecological systems to the point of collapse (Rabalias *et al.*, 2002). Intensive tilling causes soil degradation in multiple ways: decreasing soil fertility due to degradation of organic material; compacting soil from the weight of tilling

machinery which inhibits root growth, in turn, requiring more tilling; and increasing soil erosion by wind and water (Gliessman, 2000). The large amounts of nitrogen fertilizer used in industrial agriculture have their own costs: fertilizers end up in rivers, lakes and streams causing eutrophication; increased nitrogen input allows farmers to neglect the long term health and fertility of the soils; and the extraction and production of these fertilizers cause ecological damage around the world (Gliessman, 2000; Kramer et al., 2006). The most notable instance of eutrophication is the hypoxic zone known as the "Dead Spot" in the Gulf of Mexico. It is the second largest hypoxic zone in the world and a major problem for the fishing industry leading to increased mortality, reduced habitat and forced migration of many species in what used to be highly productive fisheries (Rabalias et al., 2002). Both intensive tilling and nitrogen fertilizers are used to maintain fields of monocultures, an ecological threat in their own right. Initially, monocultures decrease insect biodiversity. However, monocultures eventually cause an increase in herbivory as the insect populations become more uneven. These increases in herbivory are mitigated by pesticides which become less effective each year since insects adapt rapidly to them under the heavy selective pressure. This effect is seen in changes of herbivory from the 1950's to the 1980's, when pesticide use increased twelvefold yet crops lost to herbivory still doubled (Horwith, 1985). Sustainable agriculture, specifically intercropping, could be part of the solution to many of these problems and is currently under investigation.

### **Problems with Intercropping**

The problems with intercropping that fall within the purview of ecology stem from competitive interactions for limited resources between the crops in close proximity. Plant density plays a large role in the strength of these competitive interactions. In an experiment where peas

and barley were planted together at low and recommended densities, no significant differences in yield were found. However, when peas and barley were planted in high density, the grain yield from barley was markedly lower by the end of the growing season due to competitive exclusion by the pea plant (Hauggaard-Nielsen, 2004). Another experiment assessed the competitive interactions between cassava and legumes (Fabaceae) as well as different planting techniques. These planting techniques aim to mitigate the competitive interactions while still retaining the benefits of intercropping. In the first treatment, everything was planted at the same time, causing a 25% decrease in cassava yield due to competitive exclusion by the legumes. In the second treatment, the legumes were sown 35 days after the cassava. Tall cassava lowered legume yield by shading them. Shorter, more compact cassava had significant increases in their yield since they did not cause shading, allowing for intercropping benefits from legumes (Cenpukdee, 1992). These studies show that the negative effects of intercropping can be mitigated through farming techniques like staggered sowing times and pruning. A cost-benefit analysis of different kinds of planting techniques in the Philippines showed a fifteen to twenty year return on investment for intercropping when compared to conventional industrial methods, after which soil erosion far outweighed the greater annual cost to properly intercrop (Nelson, 1998). This shows that failing to intercrop will be more detrimental to yields and increase costs in the long run.

# Using Intercropping to Improve Soil Nitrogen Content

Though the atmosphere is composed of 80% nitrogen, it is in the diatomic form and thus inaccessible to most plants, making it a common limiting factor in ecological systems. In order for plants to use nitrogen, it must be converted into more accessible forms such as ammonium (NH<sub>4</sub>\*). Many plants do not have the ability to fix nitrogen and therefore require the presence of

other organisms to obtain nitrogen (Rascio, 2008). Legumes are one of these organisms, possessing root nodules which harbor Rhizobia bacteria. Rhizobia bacteria are able to convert atmospheric nitrogen (N2) into ammonium (NH4). The beans and bacteria form a mutualistic relationship: the beans provide a habitat and simple sugars for the bacteria in return for fixed nitrogen. Intercropping with nitrogen-fixing legumes like green beans can increase the amount of available nitrogen in the soil for partnered crops such as arugula. As Theophrastus (370-285 B.C.) stated, "... the bean best reinvigorates the soil. Beans are not a burdensome crop to the ground: they even seem to manure it, because the plant is of loose growth and rots easily" (van Kessel, 2000). There is a large base of research on intercropping relationships aiming to increase nitrogen availability, including in the replanting of forests. In Nant Port Nursery near Bangor, North Wales, an experiment tested the effect of broad and French Dwarf beans on the growth of California poplar (Populus trichocarpa). The spacing of the California poplar trees and beans was carefully assessed to ensure that the root nodules were not overcrowded, as overcrowding can cause one crop to competitively exclude the other (Hauggaard-Nielsen, 2004; Cenpukdee, 1992). The results indicated higher mean levels of nitrogen in the treatment of California poplar grown with beans as opposed to California poplar alone. Moreover, the greatest total dry weight of California poplar occurred in the treatment where the trees were placed in closest proximity to the intercropped beans. The higher mean levels of nitrogen and increased dry weight in the treatment of California poplar shows the benefits of intercropping with nitrogen-fixing beans (Ranasinghe, 1990).

Intercropping legumes with oil crops was tested recently. An experiment conducted by the Institute of Organic Agriculture assessed the effects of intercropping faba bean (*Vicia faba*) and safflower (*Carthamus tinctorius*) or white mustard (*Sinapis alba*) in the Rhineland region of

Germany. Species were grown in polycultures and monocultures. The results indicated that root density as well as nitrogen acquisition was greatest when faba beans were intercropped with oil crops. It was concluded that the varying length of the roots between the two intercropped species enhanced the use of soil-borne nitrogen and that yield of the oil crops were greater in intercropped treatments. Additionally, since the grain yield of the faba bean was reduced, the decision regarding which crop is more valuable must be determined because of competitive exclusion (Schroder, 2012).

A review of multiple agricultural practices involving nitrogen fixation in intercropping systems noted many mutualistic relationships between legume and non-legume crops. When compared to monoculture, intercropping systems had increased amounts of fixed nitrogen (specifically, faba bean and barley; and pea and barley) (van Kessel, 2000). One of the current issues with these systems is the limited amount of strains of bacteria in the soil. Inoculation of legumes with different strains of nitrogen fixing bacteria has been increasingly common in order to enhance their nitrogen-fixing potential. Often indigenous bacteria in the soil are less efficient than non-native strains due to their established adaptations to ecological conditions. Thus, they are able to reproduce in ways that do not require them to be symbiotic partners (Tajini, 2012). Therefore, research documenting the effectiveness of many strains of bacteria in different environments becomes more prevalent as intercropping is increasingly utilized.

### Intercropping as a Pest Control

A number of studies suggest that intercropping may also supplement pesticides as a form of pest control. The volatile organic compounds of some aromatic plants have been suggested to contain insect repellent properties. A study done at Beijing University of China found that

intercropping with aromatic species such as common herbs, including basil, lemon basil and savory, resulted in a significant decrease in the abundance of the beetle pest species of pear trees (Guang Bo Tang *et al.*, 2012). Additionally, intercropping with garlic has decreased green aphid populations in tobacco fields, resulting in a significant decrease in tobacco mosaic viruses transmitted by the species and an overall increase of species richness in these polycultures (Rongquan Lai *et al.*, 2011). Other studies have even reported increases in the abundance of the aphid predator species, such as the ladybeetle, when wheat fields were intercropped with garlic (Zhou Hai-bo *et al.*, 2013). These studies, and many others, stress the capabilities of aromatic plants as a viable form of biological control, using chemical ecology and plant semiochemicals such as pheromones to repel pests from important crops.

Companion plants can also act as "trap plants" to pest species. Trap plants act as diversions for herbivorous insects from their primary food source and allow for greater yields of crops as a result of this mixed planting. Intercropping brussel sprouts with imitation cereal plants promoted a significant decrease in oviposition and abundance of pest lepidopterans in a study done in the UK (David R. George *et al.*, 2013). This use of a trap species suggested that not only olfactory components but also the structural changes due to intercropping can aid in pest control. Research has even been done in order to rank oviposition and dietary preferences for certain plants by certain pests. A study done on Crambidae moths, which parasitize plants when ovipositing and consume them as larvae, attempted to rank plant preference for adult and larval moths on a number of cereal crops (Midega *et al.*, 2010). Results suggested similar plant preferences between the different life stages of the pest. Understanding plant preferences within and among pest life stages allows for efficient intercropping of trap species. Knowing there are similar preferences among life stages of a pest can reduce the number of trap species necessary

to plant and therefore prevent competition for resources. Utilizing both of these strategies, planting repellent and attractant plants, has been referred to as the push-and-pull strategy, whereby pests avoid repelling plants near the crop and are led further away by more attractive species. Such a strategy of intercropping could be beneficial for the future of sustainable agriculture.

# Arugula Experiment conducted at the University of Michigan Biological Station

Our research directly tested whether intercropping improved soil conditions and decreased herbivory. Using arugula (*Eruca sativa*) as our crop plant because of its quick growing time, we investigated the effects that green beans (*Phaseolus vulgaris*) and african marigolds (*Tagetes erecta*) had on arugula in a garden environment. Marigolds are considered a repellent of pests. Previous studies suggest that marigolds may help deter cotton bollworm from tomato plants and nematodes from cowpeas, while crushed marigold leaf and petal powder may repel deer (Srinivasan 1994, Olabiyi 2007, Abraham 1996).

We performed the experiment in the garden in at the University of Michigan Biological Station (UMBS) in Pellston, Michigan. Water sorting due to glacial activity left sandy soil in the area. Additionally, the temperate climate in Pellston makes for cool, wet springs and hot, dry summers.

#### Methods

We prepared our beds for planting by mixing Dairy Doo, a brand of cow manure, with the already present sandy soil in the garden. In planting our treatment beds, we chose to use three different beds in the garden to ensure that there were at least 10 ft. between the treatments with the marigolds and those without. It should be noted that there were 4-5 seedling transplanted at each site of an arugula plant in an attempt to decrease the probability that transplant shock or low temperatures would detrimentally affect the outcome of this experiment. For each treatment, we planted 5 replications in the same bed.

First, we planted Treatment 1 (control) which consists of arugula by itself. For each Treatment, there were 2 rows 12 in. apart from each other (Figure 1). There were 3 arugula plants in each of the 2 rows planted, 6 in. apart from one another. For Treatment 2 (arugula and beans), the arugula was again planted in 2 rows that were 12 inches apart: 3 arugula plants per row with 6 in. in between each plant. Then, between the two rows of arugula, we transplanted a row of 3 bean plants with 6 in. between one bean plant and the next (Figure 2). In Treatment 3 (arugula and marigolds), the arugula was planted in the same way as the control. For each replication of Treatment 3, there were 4 marigold flowers transplanted into small plastic pots and placed on either side of the treatment plot (Figure 3). The marigolds were planted in plastic pots in order to ensure that there was no competition for nutrients between the marigolds and the arugula plants in the garden plot. Finally, in Treatment 4 (arugula, marigolds, and beans), the arugula and beans were planted in the same way as Treatment 2. Four marigold plants in small plastic pots surrounded each plot on all four sides (Figure 4).

Four soil samples were collected from each treatment at two different times: once immediately after the plants were put in the beds and a second time after 2 weeks of growth. Soil samples were collected by hand and placed into Whirl-Paks. Each soil sample was put in a soil moisture tin and placed into an oven to dry for at least 24 hours at 60 °C. After drying, samples were processed through a 1mm sieve. Following sieving, samples were ground for at least 3 minutes. After grinding, each soil sample was placed into a scintillation vial. Samples weighing

5 grams were placed in 25 mL centrifuge tubes that had undergone an acid bath to get rid of contaminants. An extraction was then done with 25 mL of 2.0 M KCl. These extractions were shaken for 40 minutes in a shaker machine. Subsequently, extractions were analyzed by a chemist at UMBS with an auto-analyzer using standard total nitrogen methods. Values of micrograms N/g soil were then obtained.

Originally, at the end of the three weeks, we were to remove the arugula plants from their plots, dry them out, and take a total dry biomass. However, due to lack of growth as a result of cold and cloudy weather, we were unable to collect these data. Instead, we measured the area of the largest leaf of each arugula plant and counted the number of holes on each plant as a measure of herbivory. We assumed that the arugula transplanted were initially all the same starting size and leaf area. From this information, we were able to assess the amount of growth of each plant even though it was minimal, as well as the amount of herbivory in each of the treatments. For each of the 6 individual arugula plants in the 5 plots of the 4 different treatments, the length and width of the largest leaf of each arugula plant was measured. Then the number of holes on the leaves of each arugula plant were counted.

To analyze the average leaf area and herbivory level data, One-Way ANOVA tests were run. If significance was found, a Tukey's Post Hoc test indicated how treatments differed from one another.

Based on the results of the soil analysis data, One-Way ANOVA tests were run in order to determine if there were significant differences in initial and final nitrogen content. If significance was found, a Tukey's Post Hoc test was used to observe which treatments were significantly different in terms of nitrogen content. Additionally, t-tests were run between the

initial average nitrogen content and the final average nitrogen content for each treatment to observe changes of nitrogen content over time.

#### Results

Results for Leaf Area and Herbivory Level

The average leaf area did not support our first hypothesis, that intercropping legumes with arugula would increase yield, since the average leaf area for treatments with legumes were half that of the control (Table 1). The average number of holes supported our second hypothesis, that marigolds would decrease levels of herbivory (Table 1). A One-Way ANOVA test was performed for average number of holes and showed significance (p = .000; Table 2). Tukey's Post Hoc test was performed to see which groups were significantly different from others. The results of the Tukey's Post Hoc test further supported our second hypothesis since both treatments with marigolds had significantly less herbivory than those without (p = .000 in all cases; Table 3).

### Soil Sample Results

Descriptive statistics of initial average nitrogen content among treatments displayed large differences (Table 4). A One-Way ANOVA test was performed for initial average nitrogen content among treatments, showing that there was at least one significant difference in initial average nitrogen content among the treatments (p = .002; Table 5). A Tukey's Post Hoc test was performed and displayed a significantly greater initial average nitrogen content in the arugula only plot than the arugula and bean plot (p = .002, Table 6); a significantly greater initial nitrogen content in the arugula and bean plot than the arugula and marigold plot (p = .007, Table

6); and a significantly greater initial average nitrogen content at the  $\alpha$  = .08 level in the arugula and bean plot than the arugula, bean and marigold plot (p = .074, Table 6).

Descriptive statistics of final average nitrogen content among treatments displayed large differences (Table 4). A One-Way ANOVA test performed for final average nitrogen content among treatments showed that there was at least one significant difference in final average nitrogen content among treatments (p = .000, Table 7). A Tukey's Post Hoc test displayed a significantly greater final nitrogen content in the only marigolds treatment than the control treatment (p = .000 Table 8), a significantly greater final average nitrogen content in the both marigold and legume treatment than the control treatment (p = .051 Table 8), a significantly greater final average nitrogen content in the only marigolds treatment than the only legume treatment (p = .001 Table 8), and a significantly greater final average nitrogen content in the arugula and marigold treatment than the arugula, bean and marigold treatment (p = .029 Table 8). A t-test was performed on the initial average nitrogen content and final nitrogen content for each treatment. It showed a significant decrease in nitrogen content over two weeks for the arugula and bean treatment (p = .002 Table 9). An additional t-test showed a significant increase in nitrogen content over two weeks for the arugula and marigold treatment (p = .000 Table 10).

### Discussion

Leaf Area and Herbivory Level

Our first hypothesis (that leaf area would be greater in Treatments 2, 3 and 4 than in the control, Treatment 1) was unsupported by the data. This was perhaps due to plant density, starting nitrogen levels and competition for light. As previously stated, high plant density can drastically reduce yield due to competitive exclusion (Hauggaard-Nielsen, 2004). This problem

might have been exacerbated by how much more developed the beans were than the arugulas when we planted, as the bean stalks were about 6-8 inches in height as opposed to the arugulas that were less than an inch. This contradicts the farming practice of planting the superior competitor later than the inferior one to mitigate competitive exclusion (Cenpukdee, 1992). Our starting nitrogen levels were high because we tilled the soil with Dairy Doo, a highly nitrogenous compost formula (Table 4). This probably turned the legumes from a net benefit to a net negative since soil nitrogen was not a limiting factor. Finally, interspecific competition for light could have played a role since both the beans and the marigolds plants were dramatically taller than the arugula. The assumption that initial leaf area of the arugula was the same across all treatments could be another source of error. Significant differences in nitrogen content across the treatments do not seem to be an explanation for leaf area variation. This is because highest nitrogen content was found in the treatments containing arugula and legumes (which had lower leaf area than the control) while the control had the second lowest nitrogen content (and the largest leaf area).

We found highly significant differences in levels of herbivory between all the treatments with marigolds versus those without (all had p=.000; Table 3). This evidence strongly supports our hypothesis that marigolds would reduce the levels of insect herbivory present on arugula. However, there were many other plants growing simultaneously in the garden between the marigold and non-marigold treatments. Therefore, another plant may have been deterring the herbivores. A future experiment should contain two treatments, one with marigolds and one without, in a garden free of other plants that could confound the results. Additionally, the mechanisms by which marigolds deter pests should be investigated in order to determine whether it is due to their aromatic nature or the heterogeneous structure that leads to less herbivory.

### Nitrogen Level Implications

Significant differences in initial average nitrogen content among the treatments does not support our assumption that all plots started with a similar amount of accessible nitrogen. Since this assumption did not hold true, interpretation of final soil data is difficult to interpret given the initial significant differences. When mixing the soil, Dairy Doo was added due to initially high concentrations of sand that could have negatively affected soil water retention and plant growth at large. However, adding such nitrogen-rich material to the soil could have raised nitrogen levels to the point that it was no longer a limiting factor in plant growth. It is also possible that Dairy Doo was not incorporated evenly among the plots, which could explain the significant differences in initial average nitrogen content. Over time, it appeared that two of the treatments, the control and the arugula and legumes, decreased in nitrogen content. This is not surprising, considering that plants need to consume nitrogen in order to grow. However, the other two treatments appeared to increase in nitrogen content. Such results might be due to strong sampling error, whereby areas in a treatment of higher nitrogen content were sampled as opposed to areas of low content. Taking more samples per treatment might have given a better reflection of the nitrogen content.

# Improvements for Future Experiments

Unfortunately, the progress of our experiment was hampered by a lack of sunlight and warm weather during the three weeks of the Spring 2013 session at the UMBS. Since the arugula plants were transplanted from small seedlings, they were in need of optimal growing conditions in order to avoid transplant shock. The arugula transplants that successfully established were not able to produce a viable yield by the end of the three week project due to

the many evenings with a temperature around freezing, a few days of frost, and lack of photosynthesis due to minimal light during the days.

The greatest improvement to the study would be consistently warmer weather. This could be accomplished two ways: planting during the summer session or planting in a greenhouse. Future students would have to decide whether they desire more control over variables (greenhouse conditions) or increased explanatory power obtained by having exposure to the natural environment found at the UMBS (garden conditions). Either of these improvements would benefit growing conditions as well as drastically decrease the number of plants that die due to frost or other severe weather conditions. The next improvement would be to use soils of the same initial nitrogen level as well as of a lower nitrogen content. Not only do differing nitrogen levels pose a significant source of error and skew data, they also limit the explanatory power of before and after soil analysis. Finally, starting arugula from seeds instead of transplanting seedlings would eliminate error from different starting sizes as well as number of arugula plants.

### Conclusion

The research accumulated in this review as well as our recent experiment performed at the UMBS support intercropping as a substitute to the current industrial agriculture system. While our experiment did not produce ideal results due to weather complications, it can be repeated with our suggested improvements to further the understanding of intercropping and its benefits. Studies such as these not only support the use of intercropping, but also give insight to ecological systems and sustainable practices needed for the future. It is important to understand how plants interact with biotic and abiotic factors since we as humans depend on them for our

survival. Today's industrialized agricultural system is ecologically damaging and unsustainable. Based on the research found in this study, intercropping seems to be a step towards sustainable agriculture.

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# Figures and Tables

**Table 1**. Descriptive Statistics of Leaf Area and Herbivory Level (showing that the data does not support our first hypothesis at all but could support our second hypothesis)

Treatment	Average Number of Holes	Average Leaf Area (mm^2)
Control	13.0	599.8
Legumes Only	12.6	349.9
Marigolds Only	3.8	264.3
Legumes and Marigolds	1.0	250.2

**Table 2**. One-Way ANOVA Using Treatment Group as Independent Variable and Herbivory Level as Dependent Variable (Showing that there is a significant difference between at least one group)

Number of Holes	Degrees of Freedom	F-Statistic	Significance
	3	21.811	.000
Between Groups			

**Table 3**. Tukey's Post Hoc Analysing the One-Way ANOVA (showing that the significant differences lied between groups with marigolds and those without)

Treatment Groups	Significance
1-2	.996
1-3	.000
1-4	.000
2-3	.000
2-4	.000
3-4	.355

Treatment: 1= Control 2 = Beans Only 3 = Marigolds Only 4 = Both

**Table 4.** Descriptive Statistics of Nitrogen Content After Initial Planting and 2 Weeks After Planting (Displaying that our initial assumption of similar nitrogen contents in the beginning did not hold. 2 week data has not been entered yet).

Treatment	Average nitrogen content (mg of N per kilogram of soil) after initial planting	Average nitrogen content (mg of N per kilogram of soil) after 2 weeks
Control	8.425	3.7750
Legumes Only	43.125	9.100
Marigolds Only	13.425	52.600
Legumes and Marigolds	23.225	26.950

**Table 5.** One-Way Anova Test of Nitrogen Content of Initial Soil Samples Among Different Treatments (Indicating that the assumption of initial soil samples having similar nitrogen content did not hold true).

Initial Nitrogen Content	Degrees of Freedom	F-Statistic	Significance
Between Groups	3	8.901	.002

**Table 6.** Tukey's Post Hoc Analysing the One-Way ANOVA Test of Nitrogen Content of Initial Soil Samples Among Different Treatments (Indicating that nitrogen content between some treatment plots were significantly different and breaking our assumption of equal nitrogen content among all plots initially)

Treatment Groups	Significance
1-2	.002
1-3	.900
1-4	.229
2-3	.007
2-4	.074
3-4	.552

Treatment: 1= Control 2 = Beans Only 3 = Marigolds Only 4 = Both

**Table 7.** One-Way Anova Test of Nitrogen Content of Final Soil Samples Among Different Treatments (Indicating a significant difference in average final nitrogen content among different treatments).

Final Nitrogen Content	Degrees of Freedom	F-statistic	Significance
Between Groups	3	15.850	.000

**Table 8.** Tukey's Post Hoc Analysing the One-Way ANOVA Test of Nitrogen Content of Final Soil Samples Among Different Treatments (Indicating that nitrogen content between some treatment plots were significantly different)

Treatment Groups	Significance	
1-2	.902	
1-3	.000	
1-4	.051	
2-3	.001	
2-4	.157	
3-4	.029	

**Table 9.** T-test analysing the difference in nitrogen content between the initial soil samples and the final soil samples of the only legumes treatment (suggesting a significant decrease in nitrogen content of the only legume treatment over time.)

Degrees of Freedom	t-statistic	p-value
5	5.494	.002
5	regrees of Freedom	Degrees of Freedom t-statistic 5.494

**Table 10.** T-test analysing the difference in nitrogen content between initial soil samples and final soil samples of the only marigold treatment (suggesting a significant increase in nitrogen content in the marigold treatment over time).

Only Marigold Treatment	Degrees of Freedom	t-statistic	p-value
	6	-15.887	.000

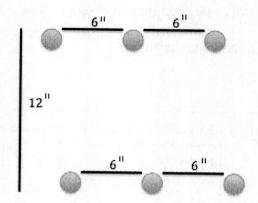


Figure 1. Treatment 1: Control

= Arugula Plant

A = Bean Plant

= Marigold Plant

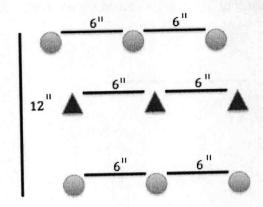


Figure 2. Treatment 2: Arugula and Beans

= Arugula Plant

A = Bean Plant

= Marigold Plant

Figure 3. Treatment 3: Arugula and Marigolds

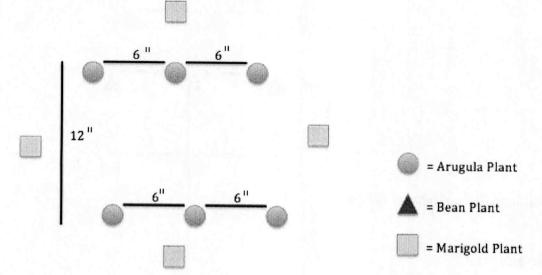


Figure 4. Treatment 4: Arugula, Beans, and Marigolds

