The Effect of Microplastics on Radish Growth

Introduction

As plastic waste grows across the world, microplastics, plastic pieces less than 5mm across, have begun to accumulate as part of the breakdown of plastic. Across the United States and Europe, it is estimated that 107,000 to 730,000 tons of microplastics are added to agricultural soils every year, while 93,000 to 236,000 tons are added to the oceans (Petersen). With so much plastic waste across the world, microplastics are being taken up by plants through their roots and spreading to many edible parts of the plant ("Research"). Microplastics may impact the growth and crop yield of the plant, a concern that may pose a threat to global agricultural production in a world of decreasing arable land and increasing population. In the past 40 years, 33% of food-producing land has been lost while current food production needs to be increased by 50% to keep up with the global food demand in 2050 (Milman). This poses a tremendous challenge in order to avoid global starvation. This project was conducted to see if microplastics significantly reduce the biomass of radish plants.

Statistical Question:

Is the biomass of radish plants grown with microplastic significantly less than the biomass of radish plants grown without microplastic?

$$H_0: \mu_{plastic} - \mu_{control} = 0 \text{ and } H_a: \mu_{plastic} - \mu_{control} < 0$$

$$\alpha = 0.05$$

Where:

$\mu_{plastic}$ = the population mean biomass of radishes grown with microplastic at the current levels in the soil

$\mu_{control}$ = the population mean biomass of the control group, the radishes grown without microplastic.

Materials and Procedure

The materials used in this experiment were plastic rubbing alcohol bottles made of Polyethylene plastic, an electric sander with sandpaper, 4 rectangular flower pots, potting soil, radish seeds, a ruler, a microwave, grow lights, a gram balance, and a scale. To prevent alcohol from entering the plants, the rubbing alcohol bottles were first thoroughly rinsed and the water was dumped out. Microplastic was then created by sanding down the plastic bottles. The electric sander was used to sand from the mouth of the bottle towards the bottom. A collecting pan was placed under the sander to collect the sanded microplastic. Most of the plastic was fine dust, however larger pieces were measured to make sure that they were less than 5mm long. Pieces that were larger than 5mm were removed from the collecting pan and thrown away.

Each pot was given the same amount of dirt as well. To measure the amount of dirt, the pot was placed on the scale and the mass without any dirt was taken. Radishes require 6 inches of soil to grow, so 6 inches of soil were added to the pot (“A Complete Guide”). This was measured by placing one end of the ruler into the pot. The dirt was poured and smoothed out until the uniform height of the dirt was 6 inches. Then the final mass of the pot with the dirt was measured. The initial mass of both pots was 1.1kg and the final mass with the dirt was 5.2kg. Thus, there was 5.2-1.1 kg = 4.1kg of dirt in each pot. According to research done by Defu He, the current amount of microplastic in agricultural fields is currently 0.54g of microplastic per kg of dirt. The pot for microplastic thus needed $0.54g \times 4.1kg = 2.2g$ of microplastic. The amount of microplastic was measured out by dumping a portion of plastic onto the gram scale. The plastic was then scraped into the pot and mixed thoroughly into the dirt. Thirty-three radish seeds were then planted in each pot. 33 was chosen as the sample size because it satisfies the central limit theorem and was the maximum number of seeds each pot could hold. Radishes were chosen for their characteristic as a cold-weather plant, as well as their ability to grow quickly. To assign seeds to the pots, the packet was shaken well and one seed was taken out. This seed was then assigned to the pot without microplastic. The packet was shaken again, another seed was dumped out and assigned into the pot with microplastic. This process of shaking the packet, taking out a seed, and placing a seed in a treatment was repeated until 33 seeds were assigned to the pot with microplastic and 33 were then assigned to the pot without microplastic. The seeds were evenly spaced and grown for 2 months in the same location and given the same amount of water. A grow light was used to give the plants light.

The method to determine the dry mass is as follows: The radishes were microwaved at 30 second intervals to remove the water. After each interval, the mass was taken using the gram scale. The radishes were microwaved until the mass of the radishes shown on the gram scale remained constant for two consecutive drying cycles. The constant mass was then recorded as the biomass of the plants.

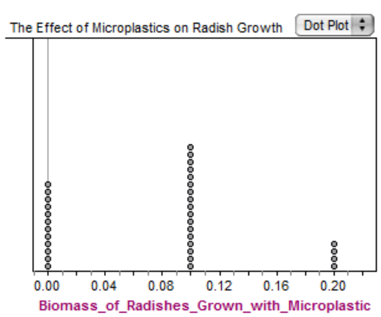
Data Display

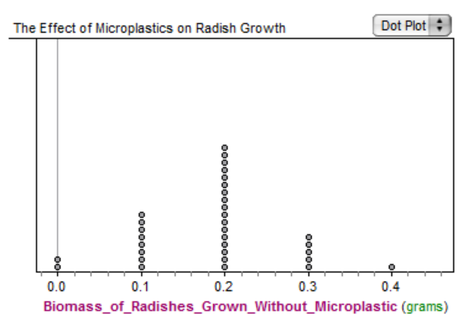
Raw Data for the Biomass of Radishes Grown with Microplastic (grams)										
0.1	0.1	0.2	0.1	0.1	0.1	0.2	0.0	0.1	0.1	0.1
0.1	0.0	0.2	0.2	0.0	0.1	0.0	0.0	0.1	0.1	0.1
0.1	0.0	0.0	0.1	0.0	0.0	0.1	0.1	0.0	0.0	0.0

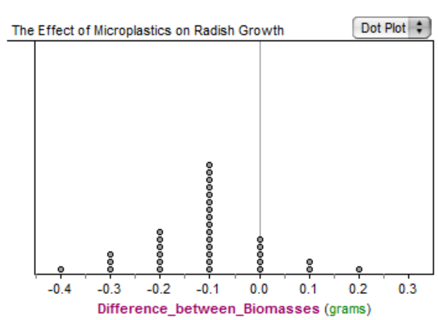
Raw Data for the Biomass of Radishes Grown without Microplastic (grams)										
0.2	0.2	0.0	0.1	0.2	0.1	0.3	0.3	0.1	0.2	0.2
0.2	0.2	0.1	0.1	0.2	0.2	0.2	0.3	0.3	0.2	0.1
0.2	0.3	0.1	0.2	0.1	0.4	0.2	0.2	0.2	0.2	0.0

Summary Statistics								
Treatment	Sample Size	Mean (g)	Standard Deviation (g)	Min (g)	Q1 (g)	Median (g)	Q3 (g)	Max (g)
Radishes Grown with Microplastic (plastic)	33	0.076	0.066	0.0	0.0	0.1	0.1	0.2
Radishes Grown without Microplastic (control)	33	0.185	0.087	0.0	0.1	0.2	0.2	0.4
Differences in Biomass (Plastic - control)	33	-0.109	0.123	-0.4	-0.2	-0.1	-0.1	0.2

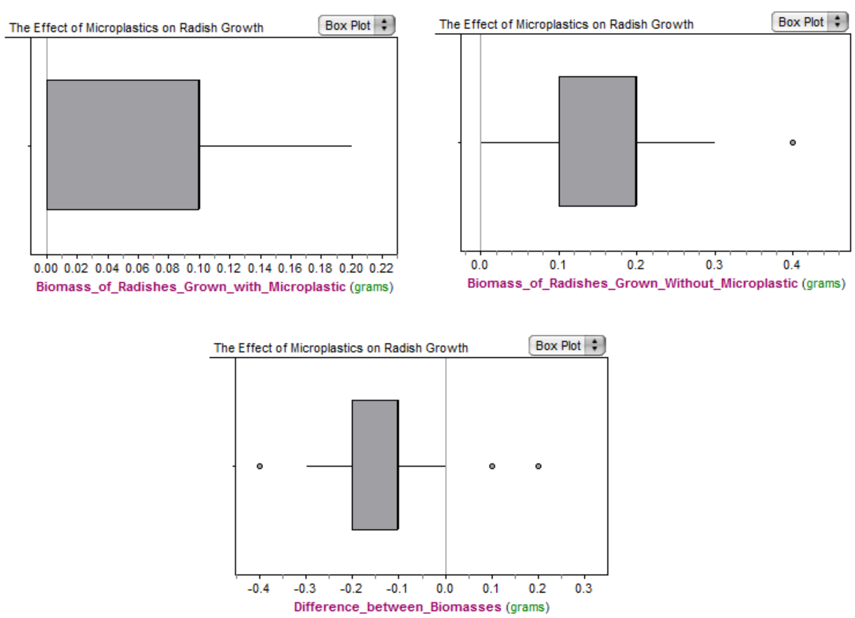
Dotplots







Boxplots



From the data display, we can see that on average, the radishes grown in microplastic are smaller than the radishes grown without microplastic. However, the radishes grown with microplastic exhibit a smaller variability than the control group. Although there seem to be outliers, all three distributions seem to be approximately normal.

Statistical Analysis

Conditions for a 2 sample t-test

1. Randomness - all seeds were randomly assigned to their treatments. First, the packet was shaken, and one seed was dumped out and placed into a pot. This was repeated until each pot had the desired amount of seeds.
2. Independence - 66 seeds out of the 1000 seed packet were chosen. 66 seeds is less than 10% of 1000, which is 100. Thus, the 10% condition is satisfied for sampling.
3. Normality - since the sample size of 33 is greater than 30, the sampling distribution of the difference of μ_{plastic} - μ_{control} is approximately normal by the central limit theorem. Although there are outliers, the central limit theorem guarantees that the sampling distribution will be approximately normal.

Outliers were determined by the 1.5*IQR rule where any outliers were above the 3rd Quartile plus (1.5*IQR) or below the 1st Quartile minus (1.5*IQR). The for the sampling distribution of the difference between biomasses, the IQR is Q3-Q1 = -0.1 - (-0.2) = 0.1, meaning that 1.5*IQR = 0.15. Thus any outliers are greater than -0.1 + 0.15 = 0.05 or smaller than -0.2 - 0.15 = -0.35. Thus the values of -0.4, 0.1, and 0.2 are outliers in this sample. However, as described above, the central limit theorem says that with a sample size of greater than 30, the sampling distribution is approximately normal. In addition, the boxplots and dotplots do not show strongly skewed data.

Calculations for a 2 sided T-test for a difference between Means		
T-statistic	Degrees of Freedom	p-value
-5.728	59.7761	<0.0001

<u>Conclusion</u>

Since the P-value of <0.0001 is less than the significance level of 0.05, the null hypothesis is rejected. There is strong evidence that the biomass of radishes grown in microplastic is less than the biomass of radishes grown without microplastic.

<u>Reflection on the Experiment</u>

In this experiment, the null hypothesis was that the addition of microplastics would not affect the growth of radish plants. However, the data and subsequent statistical analysis of this experiment showed strong evidence that the radishes grown in microplastics had a smaller mean biomass than the radishes grown without microplastic. Thus, the null hypothesis was rejected.

As a whole, this experiment went well. Every seed sprouted and produced a small radish, so that the sample size of each treatment remained the same. In addition, most of the dried radishes were able to register a reading on the gram scale, except for the very smallest ones. As a result, the data for this experiment was able to be collected and analyzed to effectively answer the statistical question.

There were several limitations in this experiment. Despite being a fast growing cold weather plant, the radishes did not grow very well. The most likely reason was because the radishes were grown indoors during the winter using grow lights, which likely contributed to the slow growth and small size of the radishes. In addition, the population means only differed by less than one gram. This tiny difference is not a practical concern for microplastics decreasing crop yields because it is so small. Therefore, the data may not have a significant real world impact, despite being statistically significant. Because the radishes were not fully grown, there is no information from this experiment on if microplastics would impact the growth of adult radishes. Further research needs to be done on how radishes would be affected by microplastics in a true agricultural setting and how microplastics would affect fully grown radishes. In addition, mean biomass for radishes grown in microplastic may have been affected because the smallest radishes were not able to register a reading on the gram scale, so the true mass would have been understated with several readings of 0.0g for some radishes.

Another limitation of this experiment was that the seeds were not planted in a true agricultural setting, but rather in small pots using grow lights. Thus, the results of this experiment can only be attributed to radish plants from the single package that was bought and grown indoors using grow lights, instead of plants grown in fields using sunlight. A potential idea for future research would be to grow plants other than radishes, such as corn or soybeans, in a field using sunlight, to see if microplastics would affect the growth of different plants in an agricultural setting.

A final limitation was that once the radishes were planted, it would have been impossible to tell if microplastic particles went into the radishes, or if the radishes were affected by plastic chemicals. The lack of a strong microscope or methods for testing chemical concentrations made it impossible to tell the difference. In addition, because the microplastics were so small, the microplastics may have been washed out through the drainage holes when the pots were watered, leaving the true amount of microplastic in the soil unknown. Another thing is that the soil purchased may already have some microplastic, further obscuring the true amount of microplastic in each pot.

Works Cited

"A Complete Guide On What Plants Can Grow In 6 Inches Of Soil." *Greenhouse Growing*, 2 Oct. 2020, www.growingreenhouse.com/what-plants-can-grow-in-6-inches-of-soil/.

Grey, Heather. "Microplastics and Your Health." *Healthline*, Healthline Media, 13 June 2019, www.healthline.com/health-news/how-dangerous-are-microplastics-to-your-health.

He, Defu, et al. "Microplastics in Soils: Analytical Methods, Pollution Characteristics and Ecological Risks." *ScienceDirect*, Elsevier B.V., 13 Oct. 2018, www.sciencedirect.com/science/article/pii/S0165993618304102.

Milman, Oliver. "Earth Has Lost a Third of Arable Land in Past 40 Years, Scientists Say." *The Guardian*, Guardian News and Media, 2 Dec. 2015, www.theguardian.com/environment/2015/dec/02/arable-land-soil-food-security-shortage.

Petersen, Kate S. "Microplastics in Farm Soils: A Growing Concern." *Environmental Health News*, Environmental Health News, 19 Nov. 2020, www.ehn.org/plastic-in-farm-soil-and-food-2647384684.html.

"Research: Crop Plants Are Taking up Microplastics." *Phys.org*, Phys.org, 13 July 2020, phys.org/news/2020-07-crop-microplastics.html.