DOI

THE EFFECT OF PH VARIATION IN IRRIGATION WATER ON THE GROWTH PERFORMANCE OF BRASSICA RAPA

Sam M. Moffit, Kara M. Scotti and Ella C. Whitman

ABSTRACT

This study aimed to investigate how varying pH levels of water affect the growth of Brassica rapa plants. Brassica rapa plants were planted in equal size containers and kept in a greenhouse where they were irrigated with equal amounts of different pH levels (3, 5, 7, 9, and 11) of water every 2-3 days. The experiment aimed to assess how pH levels influence plant growth and development and is relevant to field research of acid rainfall and mine drainage. The results revealed notable differences in growth patterns among the Brassica rapa. Beginning with germination, we noted that out of the 15 seeds planted for each pH, there were many that did not successfully germinate. Most notably were plants irrigated with alkaline water, with the pH 11 water having 10 of the 15 plants never sprout, and pH 9 which had 6 plants fail to germinate. Surprisingly, the plants watered with the very acidic water (pH 3), displayed the least number of failed plants (3). Based off our results, the pH 3 water and pH 7 water displayed the most vigorous growth, while those exposed to pH 5 and 9 exhibited slower development. As seen with failed germinations, the plants irrigated with pH 11 water exhibited signs of limited growth. Further analysis indicated that pH levels may have influenced plant's ability to absorb nutrients from the soil, impacting plant growth. The findings suggest a complex interplay between pH levels, nutrient uptake, and plant physiology in Brassica rapa plants. We noted in this study that the plants watered with pH 11 were consistently found with dry soil, despite all plants being irrigated with the same amount of water. The dry soil surrounding the plants suggests that they were not effectively retaining moisture. This could be due to increased evaporation rates due to higher pH, or hindered nutrient uptake which may have caused the plants to uptake more water.

Key Words: Brassica rapa*, pH levels, germination, imbibition*

INTRODUCTION

Water is the foundation of all life, without it, plants could not grow, animals could not live, and our planet would be just like all the others, desolate and void of life. This is because water plays a role in so many of the functions required by plants. It is the primary medium for transporting nutrients, facilitating photosynthesis, and regulating temperature within plants. Water also gives plants their structure; about 90 percent of the entire plant system is made of water, and the turgidity helps the plants stand up straight. Plants also require water as a base material for all its metabolic biochemical reactions in the plant system.

Also, water is needed to activate germination through imbibition. This process begins with a dehydrated seed, in which the cellular structures contain hydrophilic molecules. When the seed is given moisture, those hydrophilic molecules attract nearby moisture and pull it into the seed. This moisture enters the dry seed through osmosis, capillary action, and diffusion ("Imbibition Process"). Once in the seed, the structures within the seed begin to soften and swell, busting the seed casing and resulting in a sprout.

But all water is not created equally, and one way that water can differ is by pH, or the potential of Hydrogen. pH is measured using a logarithmic scale from 0 (acidic) to 14 (alkaline). The more acidic, the higher concentration of hydrogen ions there is in the solution. Rainwater is typically neutral and has a pH of around 7, so this is the pH that plants typically like. If the pH levels were to change, it could impact nutrient availability, soil structure, and the activity of beneficial microorganisms.

The pH of water that plants have access to can be impacted by various factors: pollution from industrial processes, agricultural runoff, vehicular emissions, improper waste management, and even natural disasters such as fires. These pollutants can introduce acidic or alkaline compounds into water sources, altering their pH levels. In turn, this can have detrimental impacts on plant health and ecosystem stability. One of the most well-studied examples of this is known as "acid rain," and is caused by the pollutants sulfur dioxide (SO2) and nitrogen oxides (NOx). When these harmful gas emissions react with atmospheric moisture, they form sulfuric acid and nitric acid. These acids then fall back to Earth as rain, but with a pH of less than 5.6 (Prakash et al., 2023).

Acid rain poses significant threats to plants, it can change soil chemistry, damage root systems, leach essential nutrients from the soil, and can even disrupt the natural photosynthetic processes. Additionally, acid rain can have widespread environmental impacts, including the acidification of aquatic habitats, which can harm fish and other aquatic organisms. Acid rain can also lead to the degradation of soil quality, which affects plant productivity and biodiversity.

FIELD SITE

Glaeser Greenhouse provided a warm and bright environment for the plants to grow in natural light and remain protected from harsh elements. This greenhouse was used to prevent any other confounding factor from preventing the plants from growing other than the intensity of the pH. The air inside the greenhouse is controlled to ensure there is no cross contamination from any other plant, and the experiment proceeded so that the plants reacted only to the pH-differentiated water.

METHODS AND MATERIALS

In the spring of 2024, we began initial preparations for germinating Wisconsin fast plants (*Brassica rapa)* seeds in order to conduct our experiment which studies the impact of pH on growth rates of this plant. The process started with a baking scale and 25 4-inch pots. We placed each pot on the scale, zeroed it, and subsequently filled each pot with 200 grams of moist potting soil. Following, three *Brassica rapa* seeds were spaced evenly into every pot, followed by an additional layer of soil to cover them. To initiate the germination process, 10 micropipettes of distilled water were carefully added to each pot, ensuring optimal moisture for seed activation. For ease of identification and tracking, every pot received three adhesive stickers, allowing for the precise labeling of individual seeds (e.g., 1A, 1B, 1C, 2A, 2B, 2C...).

Once the seeds were planted and watered, we brought the plants into Juniata's greenhouse, arranging them in five orderly rows within the confines of a greenhouse, ensuring uniform exposure to sunlight and temperature fluctuations. Over the course of two days, we observed the seeds germinate under the greenhouse environment. After we began to see sprouts, our focus shifted to the preparation of various water samples crucial for our investigation. This study required the creation of five distinct pH samples each which were used to water the plants throughout the experiment.

The diverse irrigation samples encompassed a spectrum of acidity, ranging from very acidic water with a pH of 3 to highly basic water with a pH of 11. The meticulous preparation of these samples was conducted within the controlled environment of a fume hood, adhering to stringent safety protocols. Crafting the acidic water involved the incremental addition of sulfuric acid to distilled water, meticulously monitored with pH papers until the desired pH was achieved. Conversely, the preparation of basic water involved a similar methodological approach, substituting sulfuric acid with ammonium hydroxide to attain the targeted pH levels.

Upon completion of sample preparation, we irrigated each pot with the water samples. We ensured that each plant received the designated pH level of water tailored to our experimental design. Plants labeled 1A through 5C received irrigation with water of pH 3, plants 6A through 10C with water that had a pH of 5, and subsequent plants were watered accordingly. While watering, every seed was watered directly with two full micropipettes, and the rest of the

DOI

sample was used to moisten the remaining soil. After watering, the water samples were stored in separate, appropriately labeled, glass containers within a cool, dark environment. Each time before the plants were watered, we used pH paper to ensure the water samples maintained the correct pH after sitting. If the pH changed as they sat, we followed the same methodology to return them back to the proper pH.

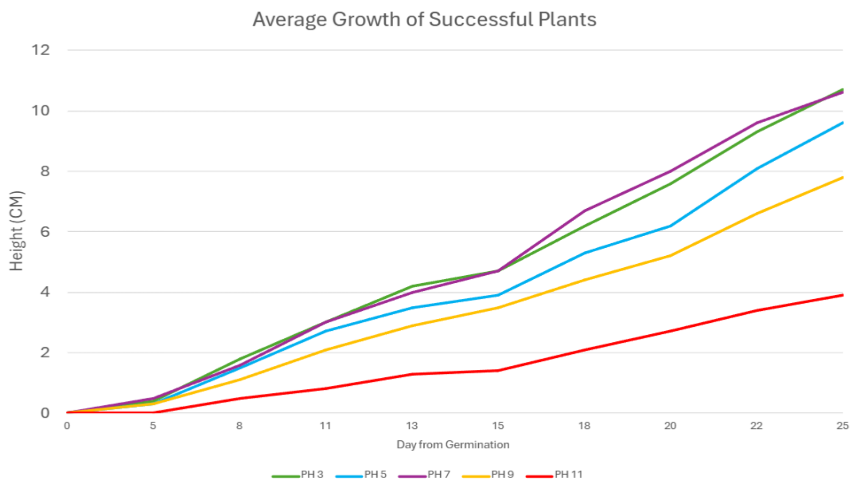
As the Brassica rapa plants began to sprout, we conducted regular monitoring, measuring, and watering every other day if possible. The greenhouse was locked on weekends, thus causing a bit of variability in days between watering and measuring. While these circumstances were not ideal, we had to adhere to Juniata's rules regarding the greenhouse. In order to measure plant growth accurately, we recorded careful measurements from the base of the plant to the tallest point using a centimeter ruler. If we saw any seedlings exhibit signs of wilting or curling, we used the ruler to stretch the seedings to full height.

RESULTS

The method used to determine the statistical analysis of the plant growth was a regression and the use of a line of best fit. Below are all the datasets entered throughout the course of the experiment. They range by color associated with each level of pH used to water the groups of plants in ascending order. The regression and line of best fit were done using the last day of research data, or data entered in the last row to the right on April 29th, 2024. The reason we chose to analyze the regression on the last day is these points are the final day the plants were measured, making them the results for the entire experiment. The inclusion of the entire data table ensures all patterns of growth are accounted for in the results.

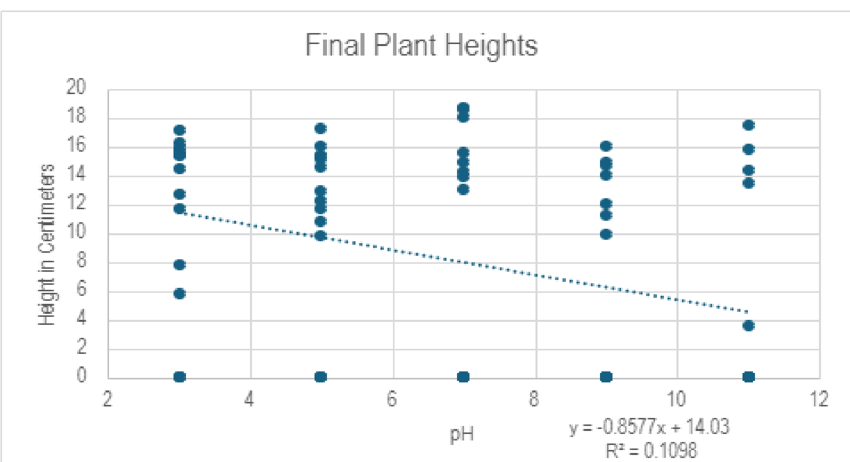
The regression line does go down throughout the course of the experiment, which means that on at least some account, the slope of the line is negative giving the regression a negative and rough slope for the correlation of plant growth height and increase in pH. Unfortunately for the hypothesis, the regression line also has an R squared value of 0.1098, which means only around 10% of the data is accounted for by the line of best fit. So, while the regression does show a slight decline in height towards the higher pHs, it shows slightly more inconsistent results than originally thought.

Without the use of the line of best fit, it is obvious that the original hypothesis was not entirely incorrect. The plant growth did peak around the pH 7 mark and stayed more consistent than any other pH. The other aspect the graph does not show are the points at which certain plants either did not grow at all or died, so they do not show up in the data but are shown on the table. There are significantly more dead or never-grown plants in the pH 11 range than in any other pH, although there is a notable amount in the pH 10 section too. This does provide sufficient evidence that the plants did have a harder time growing in the higher pHs than in the more acidic waters, as well as some of the final heights of plants that did grow are typically lower as well.



pH	Plant	Apr. 12	Apr. 15	Apr. 17	Apr. 19	Apr. 22	Apr. 24	Apr. 26	Apr. 29
3	1A	2	3.6	5	5.8	6.9	8	10.9	12.8
3	1B	1.9	2.9	3.3	3.9	5.9	6.3	7.1	7.8
3	1C	2.4	4.1	6.1	6.8	8.4	10.25	13.2	15.4
3	2A	2.6	3.8	5.2	5.5	7.2	10	12.8	14.5
3	2B	2.7	4.5	6.3	7	9.5	11.5	13.8	15.5
3	2C	3	4.8	6.6	8.5	8.6	11.5	14	15.8
3	3A	2.4	4.3	6.5	7.7	10.1	12	14.2	16.3
3	3B	0	0	0	0	0	0	0	0
3	3C	0	0	0	0	0	0	0	0
3	4A	2.1	3.8	5.7	6.5	10.1	13	15	17.2
3	4B	1.24	2.5	2.9	3	4.4	4.75	5.2	5.8
3	4C	2.3	4	4.3	5.2	7.9	11.25	14.6	16
3	5A	2.2	3.7	5.6	5.2	7.4	9.1	12.9	15.5
3	5B	0	0	0	0	0	0	0	0
3	5C	2.5	3.6	5.2	5.6	7.6	8.5	10.7	11.8
5	6A	1.5	3.4	4.2	3.7	5.6	5.75	8.8	12.3
5	6B	0	0	0	0	0	0	0	0
5	6C	0.9	0.8	0	0	0	0	0	0
5	7A	0	0	0	0	0	0	0	0
5	7B	2.5	4.1	5.9	7.1	9.5	9.8	12.7	15.2
5	7C	3.4	5	6.8	6.4	9.2	10.9	13.5	15.4
5	8A	2.6	4.5	5.6	6.6	9	11.3	13.6	16.1
5	8B	2.7	4	6	6.5	8.6	10	13.1	15.4
5	8C	1.3	2	3.4	4	5.6	7.1	8.9	9.8
5	9A	1.9	3.3	4.9	6.1	7.9	10.9	14.5	17.3
5	9B	0	0	0	0	0	0	0	0
5	9C	1.7	2.9	4.1	4.5	5.8	6.6	9.3	11.8
5	10A	1.5	3.2	4.3	5.9	6.9	8.1	12.5	13
5	10B	2.4	3.5	5.4	5.6	7.5	10.5	12.6	14.6
5	10C	1.3	3.6	4.3	6	7	7.5	9.2	10.8

7	11A	0	0	0	0	0	0	0	0
7	11B	0	0	0	0	0	0	0	0
7	11C	2.6	5.5	7	9.2	12.8	13.5	13.8	14
7	12A	0	0	0	0	0	0	0	0
7	12B	0	0	0	0	0	0	0	0
7	12C	0	0	0	0	0	0	0	0
7	13A	2	5.2	7.1	8.6	12.6	15.3	17.4	18.7
7	13B	2.2	4	5.9	7.1	11.7	13.1	16.6	18.1
7	13C	2.2	5	5.5	7.3	10.5	12.1	13.8	15
7	14A	0	0	0	0	0	0	0	0
7	14B	0.05	5.1	6.8	6.7	9.9	13.5	16	18.8
7	14C	2.3	0.1	0	0	0	0	0	0
7	15A	2.2	4.1	5.7	5.9	7.1	10	13.7	15.6
7	15B	2.1	3.6	5.2	6.2	7	7.7	11	13.1
7	15C	2.4	3.5	4.3	5.9	8.2	10.4	13	14.3
9	16A	0	0	0	0	0	0	0	0
9	16B	2.9	5.2	6.8	7.5	9.6	11.2	13.7	16.1
9	16C	2.5	4.3	5.3	6.1	8.8	10.3	12.3	14.1
9	17A	0	0	0	0	0	0	0	0
9	17B	0	0	0	0	0	0	0	0
9	17C	0	0	0	0	0	0	0	0
9	18A	2.2	4	5.7	5.9	6.8	8	9.8	11.3
9	18B	1.4	3.4	4.5	5.5	8.6	11.1	13.1	15
9	18C	0.8	2.5	3.5	5.3	5	6.9	9.3	12.1
9	19A	0	0	0	0	0	0	0	0
9	19B	0	0	0	0	0	0	0	0
9	19C	0.7	0.3	0.7	0.8	0.8	0.8	0.8	0
9	20A	1.5	3	4.4	5.7	7.1	8	11.8	14.8
9	20B	0.02	0	0	0	0	0	0	0
9	20C	1.5	2.5	4.1	5.2	6.2	6.3	8.4	10
11	21A	1.3	4	4.1	4.3	7.1	7.4	9.5	13.5
11	21B	2.2	2.1	5.2	6.2	8.1	10	13.1	15.9
11	21C	0	0	0	0	0	0	0	0
11	22A	0	0	0	0	0	0	0	0
11	22B	0.01	0.1	0.4	0.8	1.9	2	2.2	3.6
11	22C	1.92	3.2	5.1	5.4	8.7	10.3	12.9	14.4
11	23A	0	0	0	0	0	0	0	0
11	23B	0	0	0	0	0	0	0	0
11	23C	0	0	0	0	0	0	0	0
11	24A	2.2	4	5.1	5.4	7.1	10.9	14.3	17.5
11	24B	0	0	0	0	0	0	0	0
11	24C	0	0	0	0	0	0	0	0
11	25A	0	0	0	0	0	0	0	0
11	25B	0	0	0	0	0	0	0	0
11	25C	0	0	0	0	0	0	0	0



DISCUSSION

The growth of Brassica rapa plants were examined across varying pH levels, revealing insights into the influence of pH on plant development and growth. This study revealed profound implications for real-life scenarios that directly impact our environment and communities. Understanding the effects of different pH on plants can allow naturalists studying areas impacted by humans to see evidence of disruptions in the soil or rainwater pH through the impacts to vegetation around these areas. By studying how different pH levels affect the growth of Brassica rapa plants, we gain valuable insights into how acidic or basic conditions might hinder plant development.

In our study, we found that although there was variability in individual plant growth, the average collected height of all plants watered with the same pH on the same day provided a dynamic understanding of their growth rates. In this analysis, we excluded any plants that failed to germinate to avoid reducing the average height of the successfully germinated plants. Initially, all plants except those irrigated with pH 11 showed considerable growth in the first 5 days after germination. The pH 11 irrigated plants showed minimal growth, with plants barely surpassing germination height by day 7, and many of them failing to germinate altogether. This could indicate that alkaline irrigation water may have influenced imbibition differently among the plants. Plants irrigated with water of pH 11 may have experienced slower or failed imbibition due to the highly alkaline nature of the water, which can affect the permeability of cell membranes. This would impede the uptake of water by the seeds, slowing down the germination processes.

Conversely, plants irrigated with lower pH water experienced more favorable imbibition conditions, leading to quicker hydration of the seeds and activation of growth processes. As a result, these plants showed considerable growth within the first 7 days after germination. By day 8, differences in growth became evident, with plants exposed to pH 3 and 7 water exhibiting the fastest rates of growth, while those watered with pH 5 and 9 demonstrated slower development. This could be in part due to varying availability of essential nutrients at different pH levels in the irrigation water. pH levels of 3 and 7 might have facilitated optimal nutrient uptake, promoting faster growth, while pH levels of 5 and 9 might have hindered nutrient absorption, resulting in slower development. Additionally, pH levels can influence soil microbial activity, which in turn affects nutrient availability and plant growth.

Control Plants irrigated with neutral water (pH 7) exhibited the greatest final height in addition to the fastest rate of growth. This was to be expected, since the primary source of irrigation for plants in their natural habitat is rainwater, which often falls within a pH range of 6 to 7. As a result, this pH range provides the optimal conditions for plant growth and nutrient uptake. However, we did see similar rates of growth in the plants irrigated with our very acidic water (pH 3), defying the expectations set by our team. This unexpected outcome might be attributed to the adaptive capabilities of Brassica rapa plants. While acidic conditions can typically impede nutrient availability and uptake, Brassica rapa plants are fast-growing and typically used in a lab, which could lead to adaptations that make them resilient to a wide range of environmental stressors, including acidic soils.

To reach any conclusion about the effect of pH on plant growth and development, we would need to repeat this experiment on multiple other species of plants to rule out anomalies. If given the chance, further research into the underlying mechanisms affected by varying pH values could provide valuable insights for improving agricultural practices and ensuring food security in the face of changing environmental conditions.

LITERATURE CITED

eagri.org. "AGRO103 Lecture 2: Soil Physical Properties." http://www.eagri.org/eagri50/AGRO103/lec02.pdf (Accessed April 21st, 2024).

PlantPropagation.org. "Imbibition Process." PlantPropagation.org, https://plantpropagation.org/imbibition-process/ (Accessed April 22nd, 2024).

Prakash, J., Agrawal, S. B., & Agrawal, M. (2023). Global Trends of Acidity in Rainfall and Its Impact on Plants and Soil. Journal of Soil Science and Plant Nutrition, 23(1), 398-419.

doi:10.1007/s42729-022-01051-z (Accessed April 20nd, 2024).

Xu, F., Vaziriyeganeh, M., & Zwiazek, J. J. (2020). Effects of pH and Mineral Nutrition on Growth and Physiological Responses of … Seedlings in Sand Culture. Plants (Basel), 9(6), 682. doi:10.3390/plants9060682 (Accessed April 20nd, 2024).