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7 March 2020

The Effect of Sourdough Starter on Cherry Belle Radish Growth

Abstract:

Chemical fertilizers are known to have devastating effects on the environment, however the need for increased crop production to feed the Earth's growing population requires the use of growth enhancers. Sustainable bio-organic fertilizers (BOF) utilize microorganisms to induce plant growth and pathogen-resistance through bacterial colonization in the soil around plant roots, known as the rhizosphere. Certain bacteria have not been tested for their effects, such as lactobacilli and Saccharomycetales yeast strains present in sourdough starter, which would be a readily available and microorganism-rich BOF. To perform this study, 5 groups of 10 Cherry Belle radishes were watered with water-sourdough starter solutions containing 0%, 1%, 5%, 15%, and 30% concentrations of sourdough starter and were measured every two days in cm. It was hypothesized that if Cherry Belle radishes were watered with water-sourdough solutions at the above mentioned concentrations, then radishes watered with 1%-5% sourdough starter concentrations would grow the tallest and have the largest roots because beneficial lactobacillus bacteria are present in sourdough, while 15%-30% concentrations of sourdough would be too acidic for optimal plant growth. The results indicated that there was no statistically significant difference between the growth of all but the 30% sourdough concentration radish groups, which grew significantly smaller. In future studies, the microbial community present in sourdough starter needs to be further analyzed to discover its many potential uses such as a BOF, as knowledge of microbial communities is vital to the scientific community.

Introduction:

As the Earth's population grows, it's demand for food grows with it. The rise of chemical fertilizers to produce high-yield crops has proven to have devastating effects on the environment such as water pollution, soil acidification, and air pollution. Thus, a rise in bio-organic fertilizers has been observed to produce the same high-yield crops in an eco-friendly manner. Additionally, these bio-organic fertilizers have been proven to effectively reduce a plant's susceptibility to disease (NCBI, 2017). Bio-organic fertilizers function by diversifying the microorganisms present in soil, and by specifically increasing the number of beneficial bacteria. Due to the fact that soil microorganisms found near plant roots, known as the rhizosphere, strongly regulate plant growth and disease prevention (FEMS Microbiology Reviews, 2013), these microorganisms are an effective method to support plant health. Although these organisms often perform beneficial functions to the plant such as encouraging growth and acquiring nutrients, some organisms are harmful to plants, such as pathogenic bacteria and fungi. Each organism plays a specific role in maintaining or damaging a plant's health, and in order to effectively engineer bio-organic fertilizers to have a maximum positive impact on plants, it is crucial to understand which organisms are performing which functions.

Certain strains of the bacteria lactobacillus, have been shown to fight both bacterial and fungal pathogens (Hindawi, 2013). Lactobacilli are commonly found in the human gut and fermented foods such as sourdough starter, yogurt, and kimchi. These bacteria digest glucose to produce lactic acid, and some strains are proven to be probiotic organisms (AEM, 1999). The microbial communities in these systems are a simple model for the complex interactions within microbial communities. Thus, studying the microbes found in sourdough can present the scientific community with important information about the function of more complex systems such as the human gut.

The impact of microbial communities containing probiotic lactobacillus bacteria on plants has been studied in a few experiments. One study determined that cucumber growth and radish yield were improved by the addition of 90% lactobacillus bacteria, and part photosynthetic bacteria, ray fungi, and yeasts, to soil (Higa, T.; Kinjo, S., 2004). However, little research has been conducted to study the effect of fermented foods, such as sourdough, containing probiotic cultures of lactobacilli and yeast on germination rate, plant, and root growth. The sourdough industry is growing rapidly due to a global shift towards healthier and more natural products (Grand View Research, 2019). It is necessary to capitalize on this surge of sourdough products to understand the potential to recycle sourdough discard through agriculture in an attempt to reduce waste. Thus, experimentation must be conducted to understand how the addition of sourdough starter to a plant's environment impacts its growth and overall health. If sourdough starter is shown to positively affect plant health due to its large amount of lactobacillus bacteria, it has the potential to benefit the environment by both recycling sourdough discard and replacing widely used damaging chemical fertilizers. Additionally, the sourdough microbiome is a simple model for the complex subject of the interactions within microbial communities. Thus, studying the microbes found in sourdough can present the scientific community with important information about the function of more complex systems such as the human gut. It was hypothesized that if Cherry Belle radishes were watered with 0%, 1%, 5%, 15%, and 30% water-sourdough solutions, then radishes watered with 1%-5% sourdough starter concentrations would grow the tallest and have the largest roots because beneficial lactobacillus bacteria are present in sourdough, while 15%-30% concentrations of sourdough would be too acidic for optimal plant growth(Higa, T.; Kinjo, S., 2004)(Gazey, Chris, 2018)

Methods:

A mature sourdough starter that was five months old was used for this experiment. The starter was created and maintained with a one to one ratio of King Arthur all-purpose flour to water. The sourdough starter was fed every night during the experiment with 125 grams of flour and 125 grams of water. The starter was placed in a room temperature environment for fermentation every night, and was used at its peak in the morning, after it had doubled or tripled in size.

Cherry Belle radishes were then separated into five groups each containing ten radishes. Each group of radishes was planted in a large container measuring about 25' x 16' x 12', spaced two inches apart from the other radishes, and approximately ½ an inch deep. The containers were all placed in the same location next to a windowsill, thus each group received the same amount of light. They were grown at room temperature, which was maintained at about 20 degrees celsius. The ripe sourdough starter was mixed with room-temperature water at 1%, 5%, 15%, and 30% concentrations. Each group of radishes was watered with 500 grams of their respective sourdough solution. The radishes were watered every three days, and care was taken to avoid getting sourdough solution on the leaves of the plant, as the sourdough could block sunlight from reaching the leaves if it dried on the plant. Every two days, the height of each radish plant was measured in centimeters. This procedure was continued throughout a 28 day period. The results were analyzed using ANOVA and Tukey Post-Hoc Test to determine the significance ($p<0.05$) of differences in radish height between radish groups.

Results:

Minimal statistical differences in radish growth were observed. The 30% sourdough starter group did grow significantly less than the experimental groups and the control by day 20. The following shows the table of average radish height in each group over the 28 day experiment, and the visual results are shown in a graph with standard error represented by error bars.

Table 1: Average Radish Stem Height Of Radishes Watered With Varying Concentrations of Sourdough Starter Over a Period of 28 Days (cm)

		Average Radish Height (cm)														
	Day	0	2	4	6	8	10	12	14	16	18	20	22	24	26	28
	0%	0	0	0.34	4.06	6.51	6.95	7.22	7.41	7.54	7.59	7.74	7.83	7.87	7.9	7.93
Sourdough starter	1%	0	0	0.87	4.05	5.99	6.81	6.93	7.10	7.29	7.43	7.56	7.76	7.85	7.94	8.01
Concentration	5%	0	0	0.71	3.84	6.33	7.04	7.23	7.52	7.63	7.71	7.81	7.88	7.99	8.06	8.16
	15%	0	0	0.94	4.31	6.27	6.79	7.00	7.14	7.19	7.28	7.29	7.46	7.47	7.51	7.64
	30%	0	0	0.15	3.02	4.86	5.84	6.04	6.22	6.27	6.3	6.37	6.39	6.4	6.33	6.51

Table 1: *Average height of Radishes was similar for every group of radishes throughout this experiment. A notable decrease in plant height can be observed in the 30% sourdough concentration group, where the radish height on day 28 is 6.51 cm compared to the 5% group that had an average radish height of 8.16 cm.*

Figure 1: The Effect of Sourdough Starter Concentration in Water of Radish Plants on Average Radish Stem Height Over a Period of 28 Days

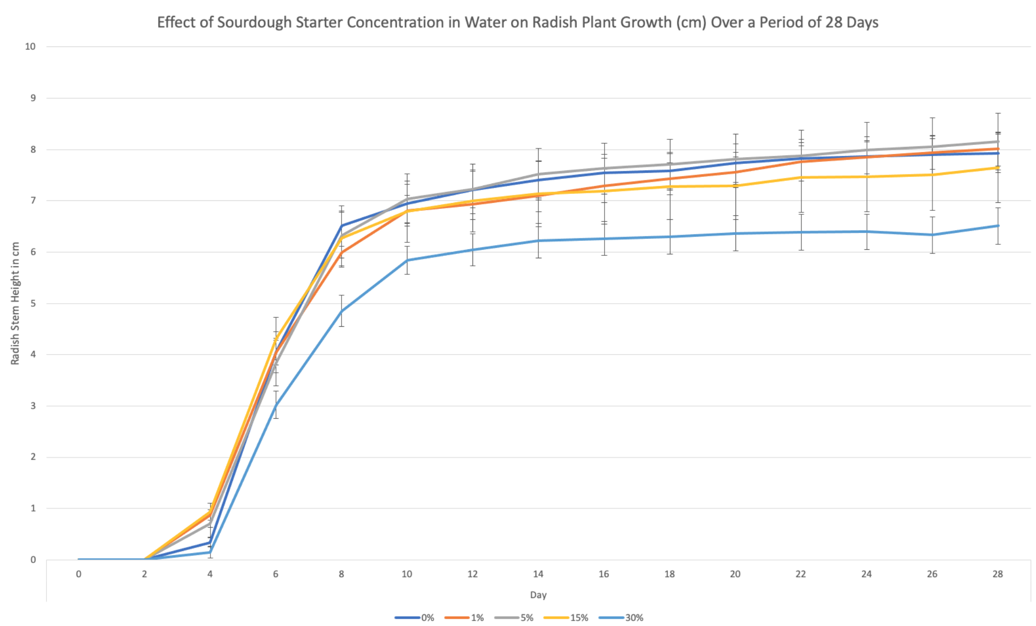

Figure 1: *Throughout the period of 28 days, the radish plants in the 0%, 1%, 5%, and 15% groups grew at a similar rate. The radish plants in the 30% group were shorter than the other plants by the end of the experiment, and error bars, which represent standard error, do not overlap from days 20-28 of the experiment, indicating statistical significance between the data.*

The radish groups showed very minimal differences in plant growth, however the data trends indicated that if the experiment were continued, the 1% and 5% groups may have grown taller than the other groups. Additionally, the 30% sourdough starter concentration group did grow less than the other groups and was supported by the lack of overlap of the error bars in figure one starting on day 20. However, when ANOVA and Post-Hoc tests were conducted, it was determined that the differences in plant growth may have been due to chance, and cannot be mathematically supported as being due to an outside factor, which would be sourdough concentration. Root length was very difficult to determine, as the plants did not produce radishes at the end of the experiment, thus the average root length was measured inaccurately, and is not included in the results of this experiment.

Discussion:

It was hypothesized that if Cherry Belle radishes were watered with 0%, 1%, 5%, 15%, and 30% water-sourdough solutions, then radishes watered with 1%-5% sourdough starter concentrations would grow the tallest and have the largest roots because beneficial lactobacillus bacteria are present in sourdough, while 15%-30% concentrations of sourdough would be too acidic for optimal plant growth. This hypothesis was rejected by the experiment because no statistically significant growth differences were observed between the 0%-15% groups. However, the hypothesis was correct in determining that the 30% group was damaging to radish plant growth. On day 28, the 30% group radishes were an average of 6.51(+/- 0.35) cm tall, while the leading 5% concentration group had an average of 8.16 cm (+/-0.55). This may have been caused by soil-surface acidity, but consistent soil pH tests registered that there was no change in the soil pH of any groups in the course of the 28 day experiment. It is possible that this occurred because the soil sample taken to measure pH used soil from varying depths and areas of the container, which may have diluted a change in soil-surface pH. The more likely explanation for the stunted growth of radishes in the 30% group is the crust that formed due to high sourdough-starter concentrations. As observed in figures 3 and 4, the 15% and 30% concentration experimental groups began to form a crust of hardened soil on top formed by dried sourdough starter. This may have caused damage to the base of the plant, as some root rot could be observed. The crust may be caused by a lack of moisture due to flour's ability to absorb water, or it could have been the result of bacterial colonization as is present in many biological soil crusts ([Warren, S.; St. Clair, L., USDA]).

Figure 3: Image of radish plants in the 15% experimental group on day 22. A noticeable crust of sourdough can be observed on the surface of the soil.

Figure 4: Image of radish plants in the 30% sourdough concentration group on day 22. A thick layer of sourdough can be seen forming a crust on the surface of the soil.

The remaining plant groups were very similar in height, however if the experiment had continued, the data trends indicate that the 5% group may have diverged in height from the rest of the group and grown more than the other groups. This would support the hypothesis, as the 1% and 5% groups were expected to grow the most. I expect this because sourdough starter contains positive lactic acid bacteria that are able to colonize sourdough starters to become the dominant bacteria species. Lactobacillus have the potential to colonize the rhizosphere of plants and prevent harmful microorganisms from growing there due to antagonistic behaviors. Seeing as lactobacillus are generally associated with pathogen resistance ([Hindawi, 2013]), their colonization of a plant's rhizosphere would generally have a positive effect on plant health because of this ability to prevent negative microbes from colonizing. Thus, one would expect that providing these bacteria would have a positive effect on plant growth, which would be supported if the 5% concentration radish growth grew taller than the other groups over a longer experiment. An experiment that may exemplify these positive effects of lactobacillus colonization of the rhizosphere may determine the effectiveness of sourdough as a treatment or prevention mechanism for common plant pathogens. Is it possible that using a direct mixture of sourdough starter and water is not the optimal way to conduct such an experiment. Perhaps centrifugation of the water-sourdough solution and use of the remaining supernatant would be better suited to optimize plant growth. This may be the case because having such a large quantity of flour in the solution is harmful to plant growth, so using the solution containing the microbial community in sourdough starter without as much flour could be vital to the success of future experiments.

Additionally, there is much to be studied on the microbiome of sourdough starters. Sourdough starters provide an optimal subject for studying simple microbial interactions within communities. As a result, greater understanding of the sourdough microbiome could lead to the ability to decipher more complex microbiomes such as that of the human gut (Cell, 2015). The microbial makeup of the specific sourdough starter used in this experiment may have also impacted radish growth. Little is known about the cause of microbial diversity within sourdough starters. Recent experiments have been conducted determining that sourdough starters are not heavily influenced by geographic location (AEM, 2020), but the type of flour used and storage conditions, as well as many other unknown factors. One such unknown is the impact of sourdough starter age on its microbiome. Future experimentation must be conducted to understand how sourdough starter microbiomes change over time, from their creation to the point of maturity. Finally, the impact of another group of bacteria, Acetic Acid Bacteria, on sourdough starter has only recently been studied and found to impact bread rise and aroma, however many other functions are yet to be uncovered (eLife, 2021). The community in sourdough starter is of high importance to understanding the microbial world. Not only might it help develop Bioorganic fertilizers that can replace harmful chemical fertilizers, but the same microbial interactions could be applied to a wide variety of subjects such as human digestion and medications; there is lots more to discover about sourdough starter.

Bibliography:

Fhoula, Imene, et al. "Diversity and Antimicrobial Properties of Lactic Acid Bacteria Isolated from Rhizosphere of Olive Trees and Desert Truffles of Tunisia." *BioMed Research International*, Hindawi Publishing Corporation, 2013, www.ncbi.nlm.nih.gov/pmc/articles/PMC3787589/.

Gazey, Chris. "Effects of Soil Acidity." *Agriculture and Food*, 2018, www.agric.wa.gov.au/soil-acidity/effects-soil-acidity?page=0%2C1#:~:text=Deficiencies%20of%20major%20plant%20nutrients,are%20less%20available%20to%20plants.&text=In%20addition%20to%20being%20chemically,root%20growth%20in%20acidic%20soils.

"Global Sourdough Market Size, Trends: Industry Analysis Report, 2025." *Global Sourdough Market Size, Trends | Industry Analysis Report, 2025*, Aug. 2019, www.grandviewresearch.com/industry-analysis/sourdough-market.

Higa, T., and S. Kinjo. *Effect of Lactic Acid Fermentation Bacteria on Plant Growth and Soil Humus Formation*. University of the Ryukyus, Okinawa, Japan, 2004, www.infrc.or.jp/knf/PDF%20KNF%20Conf%20Data/C1-5-018.pdf.

Huang, Nan, et al. "The Influence of Different Concentrations of Bio-Organic Fertilizer on Cucumber Fusarium Wilt and Soil Microflora Alterations." *PloS One*, Public Library of Science, 6 Feb. 2017, www.ncbi.nlm.nih.gov/pmc/articles/PMC5293216/.

Landis, Elizabeth A et al. "The diversity and function of sourdough starter microbiomes." *eLife* vol. 10 e61644. 26 Jan. 2021, doi:10.7554/eLife.61644

Mendes, Rodrigo, et al. "Rhizosphere Microbiome: Significance of Plant Beneficial, Plant Pathogenic, and Human Pathogenic Microorganisms." *OUP Academic*, Oxford University Press, 1 Sept. 2013, academic.oup.com/femsre/article/37/5/634/540803.

Reese AT, Madden AA, Joossens M, Lacaze G, Dunn RR. Influences of Ingredients and Bakers on the Bacteria and Fungi in Sourdough Starters and Bread. mSphere. 2020 Jan 15;5(1):e00950-19. doi: 10.1128/mSphere.00950-19. PMID: 31941818; PMCID: PMC6968659.

Reid, G. “The Scientific Basis for Probiotic Strains of Lactobacillus.” *Applied and Environmental Microbiology*, American Society for Microbiology, Sept. 1999, www.ncbi.nlm.nih.gov/pmc/articles/PMC99697/.

Wolfe BE, Dutton RJ. Fermented foods as experimentally tractable microbial ecosystems. Cell. 2015 Mar 26;161(1):49-55. doi: 10.1016/j.cell.2015.02.034. PMID: 25815984.