The Effect of pH on Yeast Fermentation Rate and Glucose Concentration

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Abstract

This experiment investigated how pH affects glucose concentration and fermentation rate for yeast. We hypothesized that a lower pH would result in a lower glucose concentration and a higher fermentation rate. Our hypothesis was refuted because the lower pH did not consistently result in both a lower glucose concentration and a higher rate of fermentation. Understanding the effects of pH on yeast is important because fermentation is widely used in the production of food and helps us understand cellular metabolism.

Introduction

Environmental conditions, such as pH, can affect how yeast use fermentation to convert glucose into energy. For example, studies have shown that yeast prefer an acidic environment and consume more glucose in low pH environments (Peña, Narendranath). Because glucose is consumed by yeast for fermentation, measuring both glucose concentration and fermentation rate can help show how yeast responds to different pH environments.

In this experiment, pH was the independent variable, with pH 5 (acidic) and pH 9 (basic) being compared as treatment groups. The control group was pH 7 (neutral). The dependent variables were glucose concentration and fermentation rate. Our research question was: How does pH affect glucose concentration and fermentation rate for yeast? We hypothesized that if the pH was lower and more acidic, then the glucose concentration would be lower, and the rate of fermentation would be higher.

Methods

Materials & Location

The experiment was performed in a standard classroom lab. We used 3 beakers of 20% glucose solutions (pH 5,7 and 9), 3 packets of yeast, Benedict's reagent, distilled water, cuvettes, microcentrifuge tubes, centrifuge, micropipettes, fermentation tubes, incubator, PCR Machine, and a spectrophotometer with accompanying software.

Procedure

We added one packet of yeast to each of the 3 glucose solutions to begin our experiment. For fermentation, we added 20 ml of each yeast-glucose mixture to the fermentation tubes, incubated them, and measured the carbon dioxide. For glucose concentration, we centrifuged 500 µl of each yeast-glucose mixture and transferred 60 µl of the supernatant into microtubes with 940 µl of distilled water for dilution. We then took 50µl of the diluted sample and mixed it with 100 µl of Benedict's reagent. The samples incubated for 5 min in the PCR machine, cooled, and then were centrifuged for 3 min. 125 µl of the resulting supernatant was transferred to a cuvette and diluted with 875 µl of distilled water. The cuvettes were put into a spectrophotometer, and the absorbance was measured at 740nm. We used the standard curve equation to calculate the glucose concentration from the raw data.

Fermentation Rates and pH Changes

	pH 5	pH 7	pH 9
Time	Fermentation Rate (ml CO_2/min)		
Week 1	0.300	0.275	0.250
Week 2	0.009	0.023	0.020
Week 3	0.007	0.007	0.007
	pH Measurements (offsets)		
Week 1	0.00	0.00	0.00
Week 2	-0.61	-2.14	-1.77
Week 3	-0.50	-2.23	-2.04

Table 1. Shows the fermentation rate at each pH level in milliliters of carbon dioxide per minute for each weekly measurement. It also shows the weekly change in pH level as an offset from the initial values of 5, 7, and 9.

Glucose Concentration

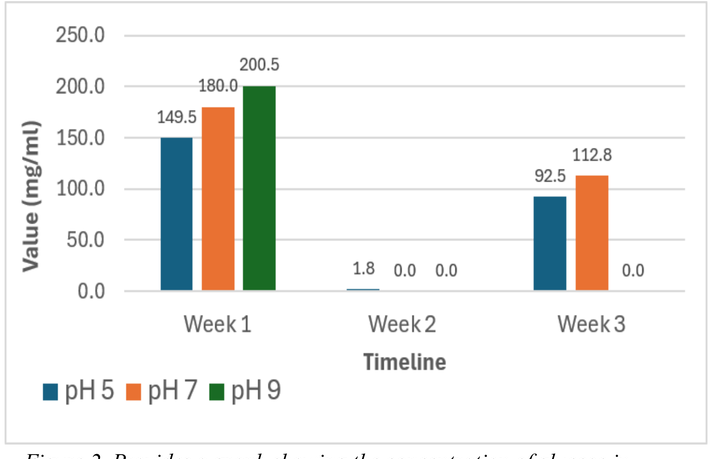

Figure 2. Provides a graph showing the concentration of glucose in milligrams per milliliter at each pH level for each weekly measurement.

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Discussion

This experiment examined the effect of pH on fermentation rate and glucose concentration over time. To test the relationships, we gathered data across 3 weeks at acidic (pH 5), basic (pH 7), and neutral (pH 7) levels. We hypothesized a lower pH would result in a higher fermentation rate and lower glucose concentration. However, the results did not support our hypotheses. The data on glucose concentration did not show linear change across the three weeks, and fermentation rate was consistent across pH levels with all dropping to almost zero by week 3, with a steep decline at week 2.

Glucose concentration was the highest at week 1 (pH 5: 149.5mg/ml, pH 7: 180.0mg/ml, pH 9: 200.5mg/ml) then decreased to almost 0 mg/ml during week 2. Week 3 showed some glucose concentration for pH 5 and pH 7, but not as high as week 1 (pH 5: 92.5mg/ml, pH 7: 112.8mg/ml, pH 9: 0mg). Week 1 results suggest that the yeast at a higher pH consumed less glucose than the yeast in an acidic pH, indicated by the high glucose concentration in pH 9. These findings align with a similar study that found yeast metabolizes and grows best in an acidic environment (Narendranath). However, a different study found the opposite, that a pH of 8 killed yeast activity (Antonio Peña). The difference between the studies was the yeast strain. The low glucose concentration detected in week 2 indicates glucose was consumed by yeast at all pH levels. By week 3, the increase in glucose concentration in pH 5 and pH 7 may indicate yeast cell metabolism or something else, whereas pH 9 still showed no glucose. Our fermentation results did not trend with glucose concentration but instead decreased over the course of the 3 weeks. Finally, the pH of each sample trended down from week 1, which could have impacted the results and changed our findings according to a similar study (Narendranath).

This experiment did not support our hypotheses. However, we were able to conclude pH has some effect on glucose consumption,. The undetectable glucose concentration in pH 9 for week 3 could suggest the yeast preferred a basic environment. However, the decrease in pH over the 3-week span may have skewed our results. For future experiments, holding the pH over time, testing different strains of yeast, or testing a larger variety of pH levels may help clarify the effects of pH on glucose concentration rates in yeast.

References/ Work Cited

1. Narendranath NV, Power R. Relationship between pH and medium dissolved solids in terms of growth and metabolism of lactobacilli and Saccharomyces cerevisiae during ethanol production. Appl Environ Microbiol. 2005 May;71(5):2239-43. doi: 10.1128/AEM.71.5.2239-2243.2005. PMID: 15870306; PMCID: PMC1087585.
2. Antonio Peña, Norma Silvia Sánchez, Helber Álvarez, Martha Calahorra, Jorge Ramírez, Effects of high medium pH on growth, metabolism and transport in *Saccharomyces cerevisiae*, *FEMS Yeast Research*, Volume 15, Issue 2, March 2015, fou005, https://doi.org/10.1093/femsyr/fou005
3. Peña, A., Cinco, G., Gómez-Puyou, A., & Tuena, M. (1972). Effect of the pH of the incubation medium on glycolysis and respiration in Saccharomyces cerevisiae. Archives of Biochemistry and Biophysics, 153(2), 413–425. https://doi.org/10.1016/0003-9861(72)90359-1