Journal of Undergraduate Biology Laboratory Investigations

2018

Sugar Rush: How Fermentation Rate Increases with Glucose Concentration

Keyla Flores, Shakota Neal, John Joseph Nguyen, Kayla Capper, Michael Felder*

[1] University of Oklahoma, Department of Biology, 730 Van Vleet Oval, Room 314 Norman, OK 73019

In the absence of oxygen, the process of fermentation is essential to a cell's ability to respire. Yeast is commonly used to produce alcohol in adult beverages through anaerobic respiration. This allows yeast to perform a modified version of glycolysis, producing NADH and ATP, along with the waste products carbon dioxide and alcohol ethanol. When yeast performs alcoholic fermentation, it uses glucose to produce ATP. The molecules available for fermentation help upkeep its ability to respire and create energy for its cells to use. These ideas led us to test differences in fermentation production. We tested our hypothesis by manipulating concentration of glucose and observing its ethanol output. Our experimental results showed strong correlation between higher input of glucose concentration and output of ethanol production. These findings supported the idea that the increased availability of organic molecules produces a higher rate of anaerobic respiration in the cell. Knowing this, we can apply the same principles back to alcohol production in order to increase the efficiency in which it is made.

Introduction

Yeast is an important staple in many goods that are produced. One such product is wine, where yeasts break down sugars in grapes to produce alcohol in beverages. (Rosini, 1984). Knowing how this process is affected by different variables helped us know how to optimize production of such goods. Cells use oxygen and glucose for respiration in order to produce ATP, which provide the cell with energy to carry out its functions. When oxygen is unavailable, glucose provides the cell with the essential molecules; carbon, hydrogen, and oxygen, to perform respiration through *glycolysis*.

Anaerobic respiration allows yeast to produce NADH and ATP, and waste products- carbon dioxide and alcohol ethanol (Shaw, 2018). The more glucose, the more ATP can be produced. One form of anaerobic respiration is alcoholic fermentation. In alcoholic fermentation, glycolysis utilizes glucose to produce pyruvic acid, which then converts to carbon dioxide and ethanol (Lin, 2006). This is the fermentation that commonly occurs in yeast, and changing the abundance of reactant variables will likely change the efficiency of fermentation within these organisms. The Crabtree effect states that higher glucose concentration increases the rate of ethanol production during

* Research Mentor

fermentation (Deken, 1966). However, other studies have come to dispute this claim by observing the activity of repression in cells that actually decrease the rate of fermentation as glucose becomes more abundant (Carlson, 1999).

To determine how yeast would actually react in practice, we collected data on how cells respire at different rates, depending on the concentration of glucose in a solution. A higher output of ethanol indicates a higher rate of cell respiration. A higher concentration of glucose will increase ethanol production because sugar molecules will be more readily available for the cell to use for fermentation. If our results supported our hypothesis, higher glucose concentration would directly correlate with a higher ethanol production. If the results did not support the hypothesis, then they showed that glucose had an adverse or neutral effect on fermentation and ethanol production would have decreased or stayed the same.

Method

To test our hypothesis that increased glucose concentration would increase the rate of fermentation, we conducted a series of tests in which we altered the proportion of glucose in a solution with yeast and measured ethanol production to determine how the change in glucose levels affected fermentation in the yeast.

First, we plugged in the stir station and prepared the ethanol probe by letting it warm up for 5 minutes. We connected the ethanol probe to Logger Pro and set up a graph to measure ppm of ethanol for every 15 seconds (2016). We set the beaker on the stir station and placed the magnet inside. Before adding the varying glucose concentrations, we added the yeast solution in the water and let it stir for five minutes at a consistent rate. For each group we manipulated the concentration of glucose to a solution with total volume of 20 mL. Group one contained an additive of 10 mL 0.3M glucose into 10 mL water and 0.6g of the prepared yeast solution. Group two contained an additive of 10 mL of 0.15M glucose into 10 mL of water with 0.6g of the yeast solution. The last group was only 10mL water and no glucose concentration, so we added another 10mL of water to conserve the 20 mL total volume and 0.6g of solution to observe the baseline. We the measured the rate of ethanol produced with each concentration. This experiment was appropriate because it allowed us to directly observe how the glucose concentration in monosaccharides affected the yeasts ability to respire. Since ethanol is an output of cellular respiration, we were able to record its rate. We compared the output of ethanol in PPM/min to the glucose concentration. The measurement of Ethanol was more suitable to the experiment because carbon dioxide is produced in aerobic and anaerobic respiration, so ethanol is specific to the fermentation. We performed a statistical analysis and did a correlational test through PAST3 spreadsheet because both types of data were numerical measurements (Hammer & Harper, 2018). We tried to determine whether there was a positive correlation between concentration of glucose and ethanol production.

Results

The rate of ethanol production tended to increase as the concentration of glucose increased, as seen in **Figure 1**. The average rate of ethanol production was 0.00135 ppm/min at 0.0 M glucose concentration added, 0.00420 ppm/min at 0.15 M, and 0.00472 at .30 M. The two groups are strongly correlated, with R= 0.81 and P= 0.0014297 meaning that there is significant correlation and very likely causality between glucose concentration and the rate of ethanol production.

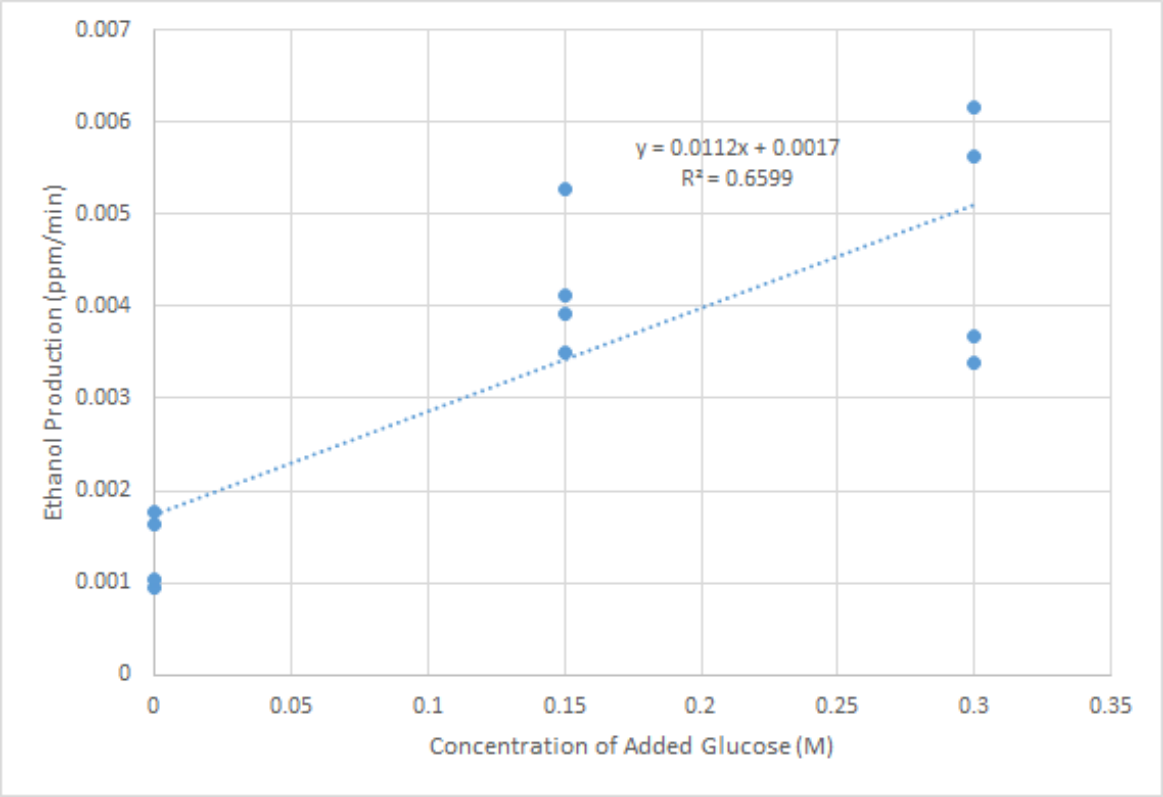

Figure 1: The rate of ethanol production for each concentration of glucose was tested over the course of 4 trials. There was an average increase in ethanol production as glucose concentration increased, with a average increase of .0112 ppm/min/M.

Discussion

The results of our experiment indicate that there is an increase in the rate of ethanol production when glucose concentration increases. This supports our initial hypothesis that a higher availability of glucose molecules to the yeast would result in an increase to the rate of fermentation within the sample. In the absence of oxygen, the yeast uses the carbohydrate, glucose to obtain energy, and ethanol becomes the product. When the amount of glucose increased, the enzymes in yeast could more efficiently ferment because of increased availability of glucose monosaccharides to the cell. Conversely, when glucose was less abundant in the solution, the yeast could not react at optimum levels, with only so much sugar reaching the cells at any one time. These results indicate that to increase the efficiency of yeast in alcoholic fermentation, producers of wine should increase the concentration of glucose in the grapes they ferment to produce more product faster. Our findings revealed how manipulating variables can change the rate of ethanol production and the concentration of sugar in the final product. The results reveal ways to create varying types of beverages. One could use a particular kind of grape with a high concentration of glucose sugar in order to produce a wine with better quality and taste. These findings could also lead to other ways of manipulating the output of anaerobic production to vary caloric content, bubbles (carbon dioxide output), and sugar (Querol, 1994).

A key flaw in this experiment is that we only took one variable into consideration when many other factors could have changed the output. We worked primarily under the assumption that it was the concentration of glucose that brought out the results, but it is possible that there were variables we did not account for. Low temperatures

are dangerous to yeast because it relaxes the uptake of amino acids, resulting in a slowing of respiration as more energy must be devoted to amino acid production (Beltran, 2007). For further research, the ability to include multiple independent variables would produce more accurate results by considering the effects of other variables: temperature, type of sugar (monosaccharide or disaccharide), water input, and time that affect anaerobic respiration.

Works Cited

Beltran, G., Rozes, N., Mas, A., & Guillamon, J. (2007). Effect of low-temperature fermentation on yeast nitrogen metabolism. World Journal of Microbiology and Biotechnology. 23(6): 809-815

Carlson M. (1999). Glucose repression in yeast. Current opinion in Microbiology. 2(2): 29-33

Deken, R. (1966). The Crabtree Effect: A Regulatory System in Yeast. Microbiology. 44: 149-156

Hammer & Harper. (2013). PAST3 (3.2) [computer software]. Oslo, Norway.

Lin Y. & Tanaka S. (2005). Ethanol fermentation from biomass resources: current state and prospects. Applied Microbiology and Biotechnology. 69(6): 627-642

LoggerPro3 (Version 3) [Computer software]. (2016). Beaverton, OR: Vernier Software & Technology

Querol A., Barrio, E., Ramón, D. (1994). Potential dynamics of natural *Saccharomyces* strains during fermentation. International Journal of Food Microbiology. 21(4): 315-323

Rosini, G. (1984). Assessment of Dominance of Added Yeast in Wine Fermentation and Origin of *Saccharomyces Cerevisae* in Wine-Making. The Journal of General and Applied Microbiology. 30(4): 249-256

Shaw, T. & French, D. (2018). Authentic Research in Introductory Biology, 2018 Ed. Fountainhead, Fort Worth.

Verstrepen, K. et al. (2004). Glucose and sucrose: hazardous fast-food for industrial yeast? Trends Biotechnol. 22(10): 531-7