JUBLI
Journal of Undergraduate Biology Laboratory Investigations

Pour Some Sugar on Me: The Relationship Between Sugar Structure and Ethanol Production, Resulting in Monosaccharides Producing a Greater Amount of Ethanol

Russell Perkins, Halle Rushton, Lauren Shephard, Tiffany Legg*
University of Oklahoma, Department of Biology, 730 Van Vleet Oval, Room 31 Norman, OK 73019

Abstract

In this experiment we aimed to understand the relationship between types of sugars and how it affects the fermentation process. We completed this experiment in order to address the question stating how variations between types of sugar, and concentrations of sugar affect the rate of yeast fermentation. While sugar type and ethanol production has been tested (Bauer et al., 2016), we addressed this question differently by exclusively testing monosaccharides and disaccharides without any sort of additives or additional independent variables. We tested this by performing a total of ten trials in which we made a solution that contained either the monosaccharide or disaccharide, yeast, and water and measuring the amount of ethanol produced as a percentage increase over a ten minute period. We hypothesize that sucrose (disaccharide) will yield higher amounts of ethanol than monosaccharide (glucose) sugars will provide more reactant material for yeast to metabolize more quickly yielding more carbon dioxide and ethanol. The procedure resulted in our data depicting a similar percentage increase between both independent variables, in addition, our Kruskal-Wallis test stated that our data was non-significant.

Introduction

The process of fermentation is vital to the creation of many popular consumable products such as soy sauce, bread, and wine. All species of yeast undergo anaerobic cellular respiration in order to produce ATP, which is necessary for cell survival(Bauer et al., 2016). The yeast consumes sugar in order to start the first step of anaerobic respiration known as glycolysis, also known as the breakdown of glucose. The yeast undergoes a series of chemical reactions during the fermentation process, ultimately yielding carbon dioxide and ethanol(Walker, G., Walker, R., 2018). Knowing that yeast feeds off of sugar in order to perform anaerobic cellular respiration, we decided to test how different types of sugar affected the metabolism of the yeast by measuring the amount of ethanol produced as a percent increase. Knowing this information could help increase the production rates of fermented commodities such as soy sauce and alcoholic beverages. A monosaccharide is the most basic form of a carbohydrate, whereas a disaccharide is simply two monosaccharides bonded together. Essentially, a disaccharide is a bigger, more concentrated version a monosaccharide(Wach et al., 2018). It is also important to point out that when too much sugar is added to a yeast concentration, there can be many negative effects on the well-being of the yeast(Delvaux et al., 2004). However, for this experiment, the moderate concentrations of glucose and sucrose will enhance fermentation performance, and cause no damage to the yeast.

We hypothesize that the disaccharide will yield higher amounts of ethanol as a percent increase than the monosaccharide because when the disaccharide bonds break, more energy is released than the monosaccharide, making the fermentation process more efficient and yielding an overall higher ethanol percentage. If our hypothesis is supported, we will see a clear positive correlation between the disaccharide and higher levels of ethanol. If our hypothesis is not supported, we will see that our monosaccharide solution yields more ethanol than our disaccharide solution, or that both solutions yield the same amount of ethanol.

Methods

In order to determine how manipulated sugar concentrations affect fermentation and its subsequent ethanol production as a percent increase the first step in our experiment was establishing a control group. The initial group established that we compared to our remaining manipulated solutions to consisted exclusively of warm water and yeast. The solution temperatures were all kept at room temperature. In addition, the temperature was kept consistent throughout the trials due to the fact that variations in temperature can affect the rate of reaction and the amount of ethanol produced. The first experimental group was a solution that was composed of warm water, 15 mL of a 0.3 M monosaccharide solution (glucose), and 0.6 grams of yeast in 15 mL of water. Once we added the 15 mL of glucose to the yeast solution we then used the ethanol sensor probe to measure the amount of ethanol produced as a percent increase over a ten minute period. Once the ten minute period concluded we simply took the total amount of ethanol produced as a percent increase. We repeated these steps, except instead of using glucose as our monosaccharide, we utilized sucrose as a our

0.3 M disaccharide.The data collected from the logger pro software was then transferred to excel where a box and whisker plot was developed in order to visually analyze the range of ethanol produced as a percent increase among the individual trials.

Results

The results were inconclusive and were not supportive of our initial hypothesis. Throughout the course of the data collection we can see that the rate of ethanol steadily increased amongst the monosaccharide and disaccharide solutions. The results were not expected due to the fact that the difference in ethanol production between the two independent variables was minimal which in turn did not allow for any valid conclusions to be drawn. In Figure 1 we can see a larger margin of difference within the disaccharides in comparison to the monosaccharide which is representative of a greater amount of ethanol produced as a percentage increase.

A Kruskal-Wallis test was conducted to compare the effect of sugar type and corresponding concentration on ethanol production on monosaccharide, disaccharide, and the control conditions. Their was not a significant effect of sugar type and and corresponding concentration on ethanol

Figures:

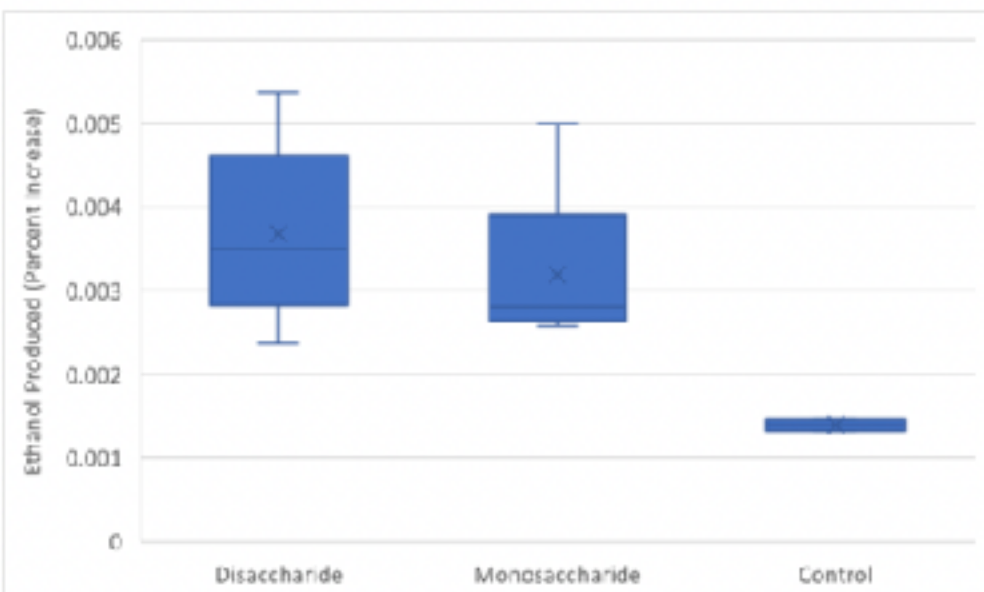

Figure 1: The box and whisker plot depicted above shows our control produced a negligible amount of ethanol whereas both the disaccharide and monosaccharide produced a measurable variance. In addition, the plot was graphed utilizing the slope of each trial which shows the rate of increase as a percent increase per minute.

production between the three conditions; [p= 0.07286]. A Dunn's post hoc test revealed ethanol production was not statistically higher in monosaccharides
(p= 0.02218) then disaccharides (p= 0.02218).

Discussion

After analyzing our results, and further research, we found that our hypothesis was not supported by our data in that glucose yielded the same amount of ethanol as sucrose. Glucose was the most efficient form of sugar during the process of fermentation, and sucrose was the second most efficient, yielding the same amount of ethanol as glucose, but more ethanol than the control group containing no added sugar. Our test did answer our initial question of which sugar structure would yield a higher ethanol percentage. Our initial thought that sucrose would yield more ethanol than glucose due to the fact that the breakdown of sucrose would release more energy, thus yielding more ethanol. After further research, we realized that we misunderstood that the process of glycolysis can only readily utilize carbohydrates in their most simple form (monosaccharide)(Moller et al., 2018). We now understand that glucose is a more effective sugar for the yeast to metabolize because it is smaller and more able to readily initiate the glycolysis process (the first step in yeast fermentation). Sucrose on the other hand, yields the same amount of ethanol, but requires energy to be converted into glucose in order to be used in the fermentation process. Because glucose by itself is able to be used directly in the process of glycolysis, it takes less energy for the yeast to metabolize and produce ethanol, while sucrose is less energy efficient when used in the process of yeast fermentation. Overall, virtually the same amount of ethanol was produced when comparing the ethanol production of both the glucose and sucrose solutions, disproving our initial hypothesis. It is important to note that many things have the ability to alter the process of yeast fermentation that we could not control in this experiment such as, temperature, osmotic pressure, oxygen, intracellular ethanol, accumulation, and yeast ethanol tolerance(D'amore 1992). These other aspects that play a part in the process of yeast fermentation could explain why our results showed an insignificant correlation between the ethanol production of glucose and sucrose when added to yeast. Overall, the information we found is important to the enhancement of the fermentation rates of future of industrial fermentation processes.

Literature Cited

Jacqueline Bauer, John Burton, Kyle Christopher, Brianna Bauer, Rachel Ritchie. (2016). Ethanol Production in Yeast According to Sugar Type. Journal of Introductory Biology Investigations. 5(2): 1-4.

D'amore, T. (1992). Cambridge prize lecture improving yeast fermentation performance. J. Inst. Brew., Vol. 98, p. 375-382.

Wach, W., Buttersack, C., & Buchholz, K. (2018). Chromatography of mono-and disaccharides on granulated pellets of hydrophobic zeolites. Journal of Chromatography. Vol 1576, p. 101-112.

Walker, G. and Walker, R. (2018). Chapter Three- Enhancing Yeast Alcoholic Fermentations. Advances in Applied Microbiology. Vol. 105, p. 87-129.

Möller. P, Liu, X., Schuster, S., Boley, D. (2018). Linear Programming model can explain respiration of fermentation products. PLOS One. Vol. 13, issue 2. P. 1-18.

Kevin J. Verstrepen, Dirk Iserentant, Philippe Malcorps, Guy Derdelinckx, Patrick Van Dijck, Joris Winderickx, Isak S. Pretorius, Johan M. Thevelein and Freddy R. Delvaux. (2004). Glucoses and sucrose: hazardous fast-food for industrial yeast?. TRENDS in Biotechnology. Vol.22 (10) 531-537.

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