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ANALYZING THE RATE OF CARBON DIOXIDE CREATED BY FERMENTATION IN YEAST WITH DIFFERENT TYPES OF SUGARS

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We researched the effects of different sugars on the rate of yeast fermentation. We were trying to discover which sugar is most desirable for alcohol fermentation in yeast. We compared water and three different sugars: glucose, sucrose, and lactose, and how they affect the rate of CO2 production in fermentation. We created solutions of sugar, water, and yeast in the respiration chamber with a CO2 probe to measure the rate of fermentation. We discovered that glucose and sucrose have a higher rate of CO2 production, due to the yeast being able to break it down into simple sugars, unlike lactose. The rate of fermentation is affected by many factors, and we use this to our advantage to make products like alcoholic beverages, yogurt, bread, and fuel in a more efficient way

Introduction

Time efficiency plays a big role in the world we are living in now. In the fermentation processes of beer glucose is used and takes about 2 weeks to complete the process. If there was a way to make the fermentation rate faster, then it would be beneficial for big beer companies, because they would save money and have a higher production rate. The rate of fermentation depends on the sugars that are used, the Crabtree Effect demonstrates that bonded molecules of glucose have lower fermentation rates than just glucose (Deken, 1965). In order to make ATP, yeast undergoes alcoholic fermentation. Yeast uses sugar to produce ATP, CO2, and ethanol (Hoefnagels, 2008). A cell can either undergo fermentation or cellular respiration to produce ATP. Cellular respiration occurs in three stages: Glycolysis, Krebs Cycle, and the Electron Transport Chain. The first stage of cellular respiration, glycolysis, utilizes glucose and converts it into 2 ATP, oxidizes NADH into NAD+, and produces pyruvate (Harris, 2015). The next step, the Krebs Cycle, or also known as the Citric Acid Cycle, occurs in the matrix of the mitochondria and cannot continue without oxygen (Weihai, 2004). The Krebs Cycle requires Pyruvate, Acetyl CoA, NAD, and FAD and produces NADH, FADH, CO2, and ATP (Weihai, 2004). After the Krebs Cycle, the Electron Transport Chain occurs in the presence of oxygen. The Electron Transport Chain occurs in the matrix, inner-membrane, and intermembrane space of mitochondria, and creates ninety percent of all ATP (Beutner, 2014). During the Electron Transport Chain NADH, FADH, and electrons create a H+ concentration gradient (Beutner, 2014). The final

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	Control (water)	Glucose (.3 M)	Sucrose (.15 M)	Lactose (.15 M)
mL of sugar	0	10	10	10
mL of water	10	0	0	0
Yeast mixture	10	10	10	10

Figure 1.1. The different experimental groups and how they are created. The yeast mixture is 10 mL of water and .6 g of yeast.

electron acceptor is oxygen, which creates the end product of water and 32 ATP (Beutner, 2014).
We tested the rate of Carbon Dioxide produced due to fermentation of yeast with different types of sugars. According to Verstrepen, yeast cells prefer glucose and sucrose when it comes to fermentation, over other carbohydrates such as lactose (Verstrepen, 2004). We analyzed sucrose, lactose, and glucose with yeast fermentation. Sucrose is a disaccharide made up of glucose and fructose with a carbon atom binding them together (NCBI, 2018). Lactose is also a disaccharide, but is composed of glucose and galactose bound together by an oxygen atom (NCBI, 2018). Glucose is a monosaccharide and is made up of an aldehyde group and six carbon atoms (NCBI, 2018). The question we tested was, does different types of sugar affect the rate of CO2 produced? We used Fleischmann's Active Dry yeast in the experiment. We hypothesize that sucrose and/or glucose will create a higher CO2 concentration over time in yeast fermentation because they have a simple chemical structure, making them easy to break down. Lactose is not as easily broken down in yeast fermentation due to yeast lacking the enzyme lactase which breaks lactose down. If our hypothesis is rejected then the amount of CO2 concentration increased more over time with lactose than with glucose.

Methods

We made the yeast according to standard protocol (Shaw and French 2018). Our study system is the yeast, we used Fleischmann's Active Dry yeast in the experiment. On the last step for the first trial we added water instead of glucose for a negative control, because the yeast would not be able to perform fermentation due to the lack of sugar and it established a baseline for the fermentation rate. We had three experimental groups; 10 mL of glucose, 10 mL of raw sucrose, 10 mL of lactose. We used these three different sugars because we were interested in analyzing the effects of different sugars on the fermentation rate in yeast, by measuring the rate of CO2 production. We analyzed the varying structures of different sugars and how they affect yeast fermentation. We constantly added 10 mL of water and .6 g of yeast in each solution, we also measure the rate of change in CO2 over a constant 10 minute period for each group. We measured the rate of CO2 production over time in ppm/min. The higher the rate of CO2 production over time, the more energy is created due to fermentation. We warmed up the CO2 probe for 5 minutes before each use. We also stirred the yeast and water for 5 minutes before adding the sugar concentrations. We measured the rate of change of CO2 by using Loggerpro and creating a linear fit, measured in ppm/min. In order to properly analyze the data trends and statistics, we performed a One-Way ANOVA. We picked this statistical test because we had nominal and measurement data, had 4 experimental groups, and had one variable.

Results

In our results, we noticed that the CO2 production increased the most in the yeast solutions with glucose and sucrose. They both had a rate of CO2 change around 1150 ppm/min, while lactose has about 456 ppm/min and water has 360 ppm/min, as seen in Figure 1.2. There are no direct outliers and the majority of the data points lie very closely together throughout the different trials. A One-Way ANOVA was conducted to compare the effect of different sugars in a solution on rate of CO2

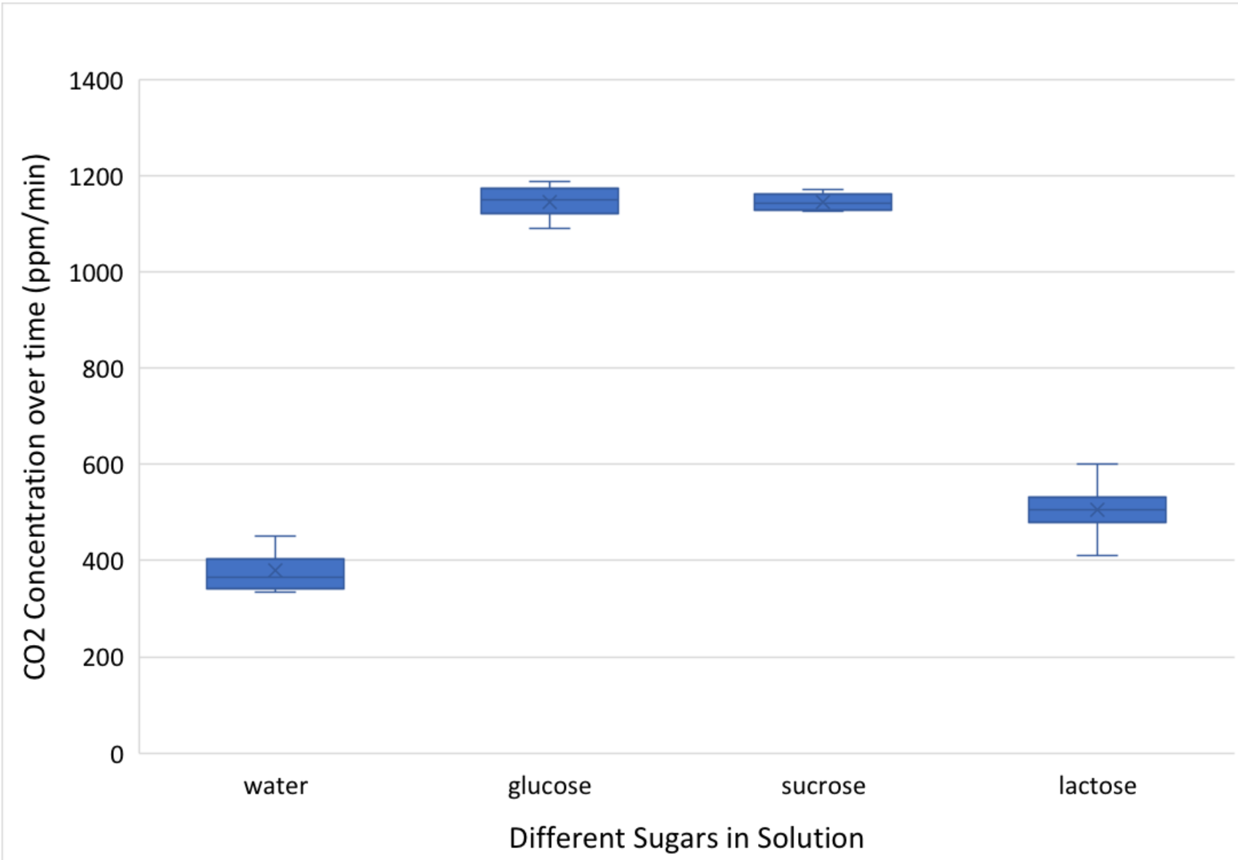

Figure 1.2. The rate of CO_2 production over time in different sugar solutions through yeast fermentation. Glucose and Sucrose had a much higher rate of CO_2 production compared to lactose and water.

production in glucose, sucrose, water, and lactose solutions. There was a significant effect of different sugars in a solution on rate of CO_2 production between the 4 conditions; [$F(3,12)= 239.4$; $p=0.0000000000573$. A Turkey's pairwise test revealed the rate of CO_2 production was statistically lower in H_2O than glucose ($p=6.341$ E-10), sucrose ($p=6.19E-10$), and lactose ($p=0.02425$). The difference between all of the solutions were significantly different other than sucrose and glucose.

Discussion

Our results show that glucose and sucrose produced the most CO_2 in ten minutes during yeast fermentation compared to lactose and water. The rate of CO_2 production increased the most with the glucose and sucrose yeast solutions than with the lactose and water, supporting our hypothesis. However, at around 8 minutes, the glucose and sucrose solutions stopped increasing in CO_2 production, around 10000 ppm. This is because the CO_2 probe had been fully saturated. We had no outliers in our data, and did not have any trends in our data that we did not expect. The only part of our experiment that could be considered unexpected was basically the same rate of CO_2 production in glucose and sucrose, even though they have different chemical structures. The product of sucrose after being broken down can be used in the glycolysis process. However, an extra step is added because fructose enters glycolysis at a step that bypasses the regulatory control exerted by phosphofructokinase, which converts fructose-6-phosphate to fructose-1,6-bisphosphate (Cason, 1987). This makes the rate of CO_2 production slightly slower. Lactose has a lower rate of CO_2 production over time due to the fact that it is a more complicated chemical structure, and is harder to break down. During fermentation sucrose and lactose are broken down into two smaller sugars. Sucrose is broken down to glucose and fructose, and lactose is broken down to glucose and galactose. However the yeast we used does not have the required enzyme, called lactase, that breaks down lactose into glucose and galactose. So when yeast is added with lactose no fermentation will occur (Science CSE, 2013). Even if lactase was added to the solution, it would only produce half of the amount of CO2 because galactose cannot be broken down by yeast in the fermentation processes. Through our investigation, we are interested in discovering how different sugars affect making products such as alcoholic beverages, yogurt, and more.

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