The Effect of Sugar Alcohols on Enzymatic Activity in Yeast Fermentation

Parnika Gupta¹

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Abstract: In this paper and experiment, I would like to explore how different sugar alcohols can affect enzymatic activity in yeast fermentation. Typically, glucose is used to activate the yeast fermentation process, and in this essay I would like to explore alternative methods to ferment yeast. This idea stemmed from my interest in fermentation, more specifically when I bake bread or create pizza dough. Fermentation has always been intriguing to me, as I find it very satisfying when I see the dough rise. Yeast fermentation is used in a variety of different products such as bread, pizza, beer, and other alcohols. It is a very common and simple process which is universally used and known. I took this idea and decided to understand more about how the process of yeast fermentation works. From there I discovered that adding sugar is necessary to the process as the sugar is the "food" needed for the yeast to activate and begin the process. This was interesting as I had recently learned about sugar alcohols in my chemistry class. This idea of using sugar alcohols instead of sugar itself to ferment yeast was alluring as I wanted to test which would ferment the fastest: the sugar or the sugar alcohol? Sugar alcohols are generally healthier than regular sugar as it is composed of a different chemical structure. This chemical structure makes it harder for the human body to digest but it allows it to contain fewer calories per gram, but still provide a sweet taste. I want to test the effect of sugar alcohols on enzymatic activity to see if it raises, lowers, or is the same as regular sugar to determine if sugar alcohols can be used in baking for the future. Sugar alcohols are a healthier alternative and I would like to put this to the test. The goal for this experiment is to help provide evidence for and support an alternative, and more healthier sugar in the fermentation process. The idea is to help those who are struggling with diabetes or are pre-diabetic, eat healthier food while still enjoying the same taste of sugar. I am pre-diabetic, and my dad was recently diagnosed with diabetes. It is difficult for him to eat food such as bread or drink wine without spiking his blood glucose levels. This study aims to test if sugar alcohols can increase carbon dioxide production, leading to higher enzymatic activity production. By identifying these sugar alcohols, this study can help develop diabetic-friendly food products that retain the same texture and taste of foods with regular white sugar. Through controlled fermentation trials using yeast, this study seeks to measure metabolic response to sugar alcohols and evaluate their impact on enzymatic activity compared to glucose.

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I. INTRODUCTION

Fermentation is the process where the central metabolism in which an organism converts a carbohydrate, such as a sugar, into an acid or an alcohol. Fermentation releases carbon dioxide gas but this is an anaerobic process, meaning fermentation occurs without oxygen. Yeast fermentation is a catabolic reaction where yeast cells break down sugars without oxygen to produce carbon dioxide, alcohol, and some ATP energy. The purpose of yeast fermentation is to "obtain energy and turn it into alcohol" (Maicas, 2020). This process of producing gases, acids, or alcohols are carried out by enzymes.

Enzymes are biological catalysts or globular proteins that promote the rate of a specific reaction in a cell by decreasing the activation energy for the process. Enzymes are catalysts, so they reduce the activation energy needed to create the products. They are crucial in the metabolic pathways of anabolic and catholic reactions. Enzymes are specific, as they only bind to the substrate that fits into their

active site. Substrates require a specific enzyme to catalyze their reaction and only the correct substrate is able to bind to an enzyme. After the reaction occurs, the enzyme can be used again and can bind with a new substrate.

Sugars are a type of carbohydrates, specifically monosaccharides and disaccharides. Monosaccharides like glucose and fructose, and disaccharides like sucrose and lactose are types of sugars that are commonly found. The most common sugar that we use is sucrose, and it is a disaccharide formed of glucose and fructose that is naturally made in all plants. "Glucose is the fuel the brain, organs, and muscles need to function and engage in everyday activities" (The Sugar Association, 2019). Glucose is the simplest and most abundant monosaccharide, mainly made by plants and algae during photosynthesis.

Sugar alcohols, also called polyols, are a type of carbohydrate whose structure resembles both sugars and alcohol" (Mandl, 2020). Sugar alcohols are similar to sugar as they have the same sweet taste when consumed, and have

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the same cooling, pleasing effect like regular sugars. However, their chemical structure is different from regular sugars so they are not easily digested or absorbed. Sugar alcohols contain a hydroxide group, which means they are sweet like sugar but are not metabolized in the same way. The benefits of sugar alcohols compared to sugars are vast, and better for the majority of people who enjoy the taste of sugar. Sugar alcohols contain fewer calories per gram than regular sugars as on average they supply around two calories per gram compared to sugar's four calories per gram. However, the human body does not digest sugar alcohols efficiently, and excessive amounts of it can lead to gastrointestinal problems like abdominal pain, diarrhea, or cramps. Since sugar alcohols are slowly metabolized, this can lead to development of gut bacteria and production of excess gas. Sugar alcohols do not have as prominent of an effect on blood glucose levels than sugars do. Since sugar alcohols are not easily absorbed, this can lead to prevention of spikes in blood sugars, and this can be a useful alternative to people who are diabetic or pre-diabetic.

II. METHODS

Hypothesis

Sorbitol has a prominent effect on Carbon dioxide levels in yeast fermentation as it increases the number of reactions compared to other sugar alcohols like xylitol and glycerol because of its molecular structure.

➤ Variables

 Independent Variables: Xylitol, Sorbitol, Mannitol, Glycerol

- Control: Glucose
- Dependent Variable: CO₂ concentration (ppm/sec)
- Controlled Variables: Sugar concentration, solution volume, yeast amount, fermentation time, temperature

➤ Materials

- Instant baker's yeast
- · Distilled water
- Sugar alcohols (Sorbitol, Xylitol, Glycerol, Mannitol)
- Glucose
- CO₂ gas sensor/probe
- Thermometer, balance beam, graduated cylinders, beakers
- Hot plate, stirring rod, sensor chamber

➤ Procedure

Solutions of 10% concentration were created for each sugar alcohol and glucose by dissolving 30g of substance in 300mL of 37°C water. A 4% yeast solution was prepared by dissolving 10g of yeast in 250mL water. For each trial, 90mL of sugar solution and 10mL of yeast solution were combined in a gas chamber and sealed with the CO₂ probe. Each sugar was tested in three trials for accuracy.

III. RESULTS

Xylitol released the most CO₂, suggesting the highest enzymatic activity. Data was graphed using Excel to illustrate differences between sugar alcohols. Xylitol outperformed all others, followed by mannitol, sorbitol, glycerol, and finally glucose.

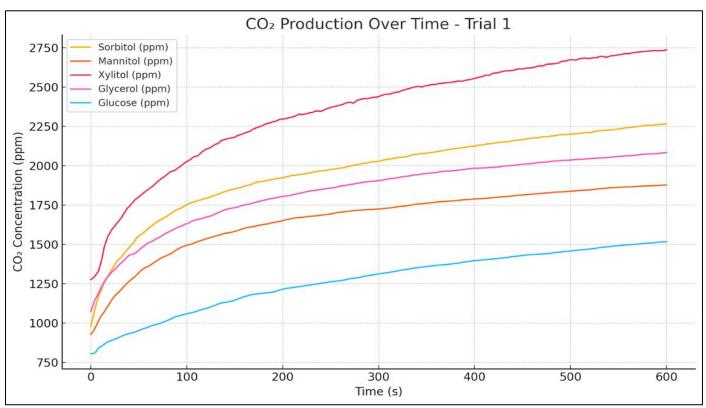


Fig 1 CO₂ Production Over Time - Trial 1

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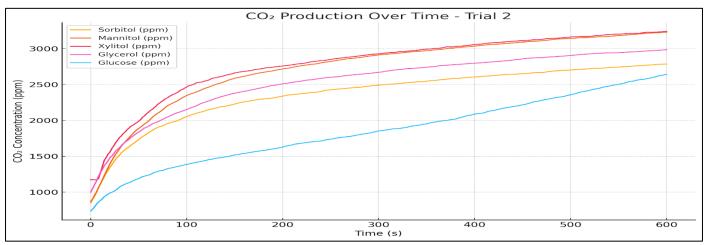


Fig 2 CO₂ Production Over Time - Trial 2

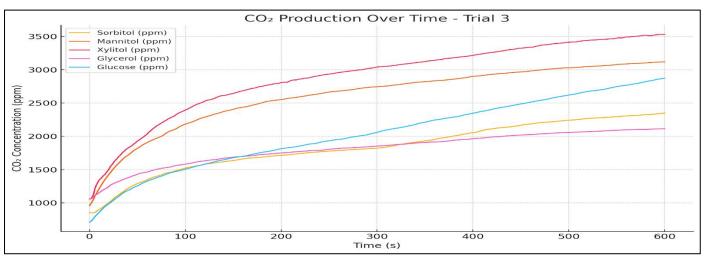


Fig 3 CO₂ Production Over Time - Trial 2

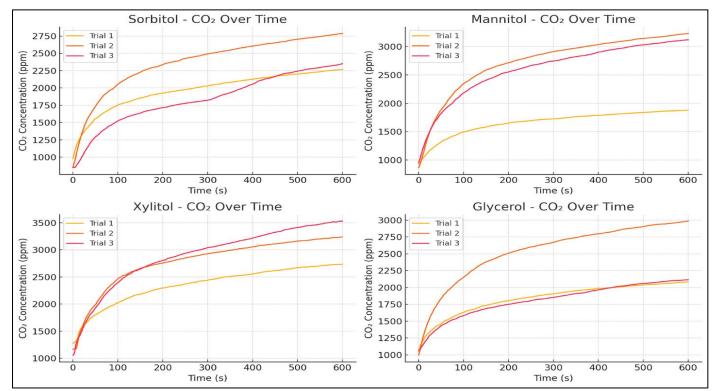


Fig 4 CO₂ Over Time

IV. DISCUSSION

Using Figures 1, 2, and 3, xylitol produced the most carbon dioxide indicating that the most enzymatic activity occurred when the xylitol solution was added to the yeast. This shows that xylitol had the most enzymatic activity, contradictory to my hypothesis. I originally hypothesized that glycerol would release the most carbon dioxide because of its small size and symmetry. Glycerol would easily fit into the active sites of the enzymes. In addition to this, I also hypothesized that glucose would release the most carbon dioxide because of its ability to digest easily, and its simple monosaccharide structure. The results were unexpected, and this could have happened because of a few errors in the lab or due to the yeast being stressed. Xylitol produced the most carbon dioxide which typically goes against the predictions. This could be the case for a numerous number of reasons, but the main reason could be that the yeast contains the enzyme specific to Xylitol called xylitol dehydrogenase. Mannitol and sorbitol both produced moderate amounts of carbon dioxide as these sugar alcohols are partially fermentable. A reason to suggest why this is happening is because immediately after the initial lag phase, the enzymes are converting the sugar alcohols into glycolysis intermediates. Glycerol surpassed glucose, but it was expected to release the greatest amounts of carbon dioxide. Its output suggests that yeast were capable of slow metabolic conversions.

In this experiment, glucose resulted in the lowest carbon dioxide output, indicating the lowest enzymatic activity in yeast fermentation, whereas xylitol produced the highest carbon dioxide output. This contradicts the standard fermentation process as glucose is expected to be the most fermentable substrate. These results suggest that either the yeast strain used adapted to metabolize sugar alcohols more efficiently, or that experimental or environmental factors influenced the fermentation differently than expected. Examples of these factors would be temperature differences, pH, or inconsistencies in the solutions prepared for glucose. For instance, while the glucose trials were conducted at 25 degrees Celsius, some of the other sugar alcohol trials were conducted at higher temperatures such as 27-31 degrees Celsius. This could have enhanced the enzyme activity and fermentation rates as temperature can change the behavior of an enzyme.

Conversely xylitol, which is generally regarded as poorly fermentable, produced the most carbon dioxide. Factors such as minor differences in yeast solution volume, sugar solution volume, temperature variety, and even the purity of the sugar alcohol could have influenced the enzyme-substrate interaction. Mannitol and sorbitol showed intermediate results, which was consistent with the potential for partial metabolism after conversion to glycolysis intermediates. While glycerol is known as a non-fermentable substrate under anaerobic conditions it was still relatively low but better than glucose. The initial data highlights the importance of experimental consistency and the sensitivity to environmental variables. These unexpected findings suggest a need for a deeper evaluation of the yeast strain used with more tightly controlled parameters.

V. CONCLUSION

The purpose of this essay was to analyze how different sugar alcohols such as glycerol and mannitol, would affect how much carbon dioxide product was released, and using this information, analyze the differences in enzymatic activity during the fermentation process. Contrary to predicted results, glycerol and glucose resulted in the lowest carbon dioxide production, suggesting that the enzymatic activity was limited in those solutions. Surprisingly, xylitol showed the highest levels of carbon dioxide output which indicated unexpectedly high levels of fermentation, suggesting possible adaptations or different yeast strains. The pre-lab data revealed that while the sugar concentrations and sugar volumes were carefully controlled and created, there were still variations in the temperature and in the solutions created. These results emphasized the complexity of yeast fermentation and metabolism as there are a variety explanations for the differences and inconsistencies in results. Future investigations could benefit from increased control over the variables such as temperature and pH, use of better technologies like an incubator or stronger carbon dioxide probe, or even yeast strain verification. Overall, this experiment provides compelling evidence that sugar alcohols, in particular xylitol, can significantly influence fermentation rates and how they function. This study suggests the need for further biological and biochemical interactions with yeast.

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