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Investigating The Fermentation of Yeast

I am going to investigate the fermentation of yeast with the use of liquid glucose as food.

Background on Yeast

Yeast cells are microscopic, one-celled fungi which are important for their ability to ferment carbohydrates in various substances. Yeasts are widespread in nature. They are found in the soil and on plants. Most yeasts used by man belong to the branch of living organisms called *Saccharomyces*. Those known as brewers' yeasts are strains of *Saccharomyces cerevisiae*.

Yeasts are well known for the making of bread and wine. Today they are used industrially in a large range of fermentation processes;

- medically, as a source of B-complex vitamins and thiamine
- as a stage in the production of various antibiotics and steroid hormones
- as animal feed and foodstuff for humans.

A yeast-like fungus is a source of riboflavin (vitamin B2) which is good for the skin. Fungi are also used to produce Roquefort cheese and to ripen Camembert cheese.

Yeast cultures are grown in a medium of sugars, nitrogen sources, minerals, and water. The final product may take the form of dried yeast cells, or the yeast may be pressed into cakes with some starchy material. When a batch of yeast for baking, medicinal, or food purposes is made, the stuff in which the yeast was grown is thrown away. In the making of wines, beers, spirits and industrial alcohol, however, it is the fermented liquid which you want and the yeast itself is waste or is used for animal feed. Fermentation of glucose by yeast produces ethyl alcohol and carbon dioxide.

Some Information On Glucose

Glucose, or monosaccharide sugar as it is sometimes known, has the chemical composition $C_6H_{12}O_6$. You find it in honey and the juices of many fruits; the alternative name *grape sugar* comes from the presence of glucose in grapes. Glucose is a normal constituent of the blood of animals.

Glucose is a white crystalline solid but is less sweet than ordinary table sugar. Glucose is formed from many carbohydrates, including sucrose, maltose, cellulose, starch, and glycogen. Glucose is made industrially by the hydrolysis of starch under the influence of dilute acid or, more commonly, under that of enzymes. It is chiefly used as a sweetening agent in the food-processing industries. It is also used in tanning, in dye baths, and in medicine for treating dehydration and for intravenous feeding.

Explanation of the experiment

I am going to set up a simple experiment to see how the fermentation of yeast develops at room temperature. I am going to make use of the fact, already mentioned above, that when fermentation takes place a gas, Carbon Dioxide (CO₂), is given off as a waste product. When the fermentation is strong more gas will be made than when the fermentation is weak. I do not need to know exactly how much gas is produced - I will only need to be able to judge if more or less is being given off.

Because there is limited time in the lesson for the experiment my partner and I are going to do two at once and so take two sets of results at the same time.

I think there are four variables I could be dealing with:

- the temperature the mixture is kept at,
- the amount of yeast used,
- the amount of glucose used,
- whether the mixture is in the dark or the light.

Investigating the Temperature

I am going to set up an experiment where I have two test tubes both containing three ml's of glucose solution and three ml's of yeast mixture. The yeast I am going to be using is dried brewers' yeast mixed with water and the glucose too is mixed with water. I am then going to put two rubber bungs, each of which have small hollow glass rods going through them, into the top of the two test tubes. Two rubber tubes are then going to be fixed around the top of the two glass rods. The other ends of the rubber tubes are going to be placed into two other test tubes filled with water. As the yeast ferments the yeast gives off carbon dioxide. The carbon dioxide then builds up and to release the pressure, gets forced out of the end of the rubber tube. I shall then count the bubbles and base the rate of fermentation on the results. I shall count the bubbles over a period of one minute and do this for twenty minutes.

Next, I shall repeat the experiment a number of times using a mixture which is starts at higher temperatures and see what effect this has on things.

Each time I do the experiment I am going to use two test tubes of mixture to try to even out any odd things that might happen.

Here is a diagram of my experiment:

What I think will happen

I expect that to begin with there shall only be a small amount activity from the yeast for there will not have been enough time for the growth of yeast cells. But, as time goes on, more yeast will begin to build up by feeding on the glucose solution and so more gas shall be given off. As the experiment continues the glucose will be used up and this will be shown by the amount of bubbles being given off slowing down.

As I raise the temperature at which the experiment takes place I expect the fermentation to increase because the heat energy will encourage the yeast to reproduce at a faster rate. I will be able to tell this is happening by a greater amount of bubbles of CO₂ being given off.

Experiment one

Here are my results from this first trial:

TIME (minutes)	Number of bubbles given off /min.	
	TEST TUBE ONE	TEST TUBE TWO
1	0	0
2	0	0
3	0	0
4	0	0
5	0	0
6	0	0
7	0	0
8	0	0
9	0	0
10	0	0
11	0	0
12	0	0
13	0	0
14	0	0
15	0	0
16	0	0
17	0	0
18	0	0
19	0	0
20	0	0

I am not going to put these results into a graph because there is no point for it would only give a very uninteresting straight line.

Whilst doing the experiment I noticed a build up of a thick creamy coloured cloud in the bottom of the test tubes. I was told that this was the yeast developing in the bottom of the test tube. I could also see froth above the surface of the liquid. This indicated that the yeast was starting to ferment and give off Carbon Dioxide.

By looking at my results it appears that my prediction was wrong. The yeast did take some time to start to ferment but in the twenty minutes of the experiment not one bubble emerged from the tube.

I think this may have been caused by excess pressure in the rubber tubes, for the ends of the two tubes were under more water than was necessary. Another reason for these results may be that the yeast was too cold in the test tubes to ferment well. Also the ends of the rubber tubes are quite a large diameter needing quite a lot of gas build-up before any bubbles are given off.

Experiment two

To see if the tube diameter and pressure were the cause for the lack of bubbles, I shall now do a similar experiment but I shall use some special glass tubes to go into the water. These fit into the rubber tubing and taper at the other end to a small diameter hole. This should make it easier for the bubbles to be seen because it will only take a small amount of gas to make them. I am also going to reduce the depth of the water the tubes are placed in to reduce the pressure. This will hopefully allow the bubbles to flow out with more ease.

I predict that with these changes to the original experiment I shall record at least some bubbles per minute.

Here is a diagram of the experiment:

Here are the results of experiment two in a table:

TIME IN MINUTES	Number of bubbles given off /min.	
	TEST TUBE ONE	TEST TUBE TWO
1	0	0
2	0	0
3	0	0
4	0	0
5	0	0
6	0	0
7	0	0
8	0	0
9	0	0
10	0	0
11	0	0
12	0	0
13	0	0
14	0	0
15	0	0
16	1	1
17	1	1
18	2	0
19	3	2
20	3	2

My prediction was correct. This time some bubbles were counted although not many. I am going to graph these results because I think that these will do as my first set of results.

It took the yeast sixteen minutes before the first bubble came out of the rubber tube. I was surprised that it took so long for a bubble to come out of the rubber tube because I saw froth above the liquid's surface in both test tubes after a few minutes indicating that the yeast were fermenting.

Experiment three

I believe that the results of experiment two are not very dramatic because the yeast was too cold. I shall redo the experiment putting the test tubes holding the yeast/glucose solution in a beaker of warm water to see if this helps. I shall do this by boiling a kettle and pouring the 50mls of hot water into a beaker. I shall then add 50mls of cold tap water to it, cooling the temperature a bit. I shall still use the same amount of yeast and glucose as I have in the previous two experiments to keep the tests fair.

I predict that with these changes to the original experiment I shall get a good set of results because with the warm temperature the yeast shall reproduce at a faster rate.

Here is a diagram of this next experiment:

I am now going to put the results into a graph and a table.

TIME IN MINUTES	Number of bubbles given off /min.	
	TEST TUBE ONE	TEST TUBE TWO
1	7	6
2	5	5
3	2	1
4	1	4
5	1	1
6	2	2
7	1	1
8	1	2
9	1	1
10	3	1
11	2	2
12	2	1
13	1	1
14	2	1
15	2	2
16	1	1
17	1	2
18	1	1
19	1	2
20	2	1

My prediction was correct. The changes that were made did help with the fermentation of the yeast. The yeast under these conditions gave off quite a steady stream of bubbles indicating that the fermentation had been helped by the increase in the temperature of the mixture. I am pleased with this result. By looking at the table and the graph I noticed that as time went on the amount of bubbles given off by both of the tubes slowed down. I think this may have been caused by the drop in the water's temperature as time went on as the heat was radiated away, or the yeast may have used up the glucose and slowed the fermenting right down.

Experiment four

I was only able to do one more experiment. I put very hot water, again from the kettle, into the beaker surrounding the two test tubes without adding any cool water to reduce the heat at all. This gives the yeast a lot more heat which should help the fermentation process even more.

Here is a diagram of the experiment:

I predict that with this hotter temperature the yeast will ferment at a very quick rate though this will slow down as time goes on because the water temperature shall cool and as more yeast is produced the food shall run out.

I am now going to put the results into a graph and a table.

TIME IN MINUTES	Number of bubbles given off /min.	
	TEST TUBE ONE	TEST TUBE TWO
1	24	21
2	19	16
3	5	6
4	0	4
5	1	0
6	0	0
7	0	0
8	0	0
9	0	0
10	0	0
11	0	0
12	0	0
13	0	0
14	0	0
15	0	0
16	0	0
17	0	0
18	0	0
19	0	0
20	0	0

As soon as I put the test tubes into the water I had bubbles coming out of both glass rods straight away. One tube gave off lots of bubbles but the other, for some reason didn't give off even half as much. At a guess I would say that the actual content of yeast in the yeast/water solution was greater in test tube one than in test tube two. My prediction was correct. Yes the yeast did ferment at a much greater rate than before. And yes this did slow down as time goes on. In actual fact both test tubes had stopped giving off bubbles after the fourth minute. I did not think that this would happen but it did. I think the main cause for this was the temperature was too hot. The only reason I can think of why the yeast was not killed immediately is because it took a while for the heat to conduct through the glass test tubes and heat up the solution.

By doing these experiments I found that yeast need a continuous supply of glucose and a constant heat to ferment as well as they can. I do not think my results were as accurate as possible even though I tried to create conditions that were the same for the experiments to create a 'fair test'. The problems that occurred during the experiments were as follows;

- I could not control the amount of bacteria that were in the yeast/water solution in each of the test tubes.
- I could not keep the test tubes at a constant heat.
- I could not keep feeding glucose into the test tubes without taking out some of the yeast/glucose mixture.
- I could not make sure that there were equal amounts of pressure on each rubber tube/glass rod.
- I only had a certain amount of time in which I could only do a certain amount of experiments.
- I would have liked to have taken the temperature at each interval when I recorded the amount of bubbles given off by the two test tubes but I didn't.

If I was to do the experiments again I would take more care in controlling the problems that I had this time.

Because of the problems I experienced and lack of results, I do not think I can come to a firm conclusion, though by doing some research I can write down the equation to show what happens when yeast ferments, showing what they need to ferment and what the waste products are (shown on next page).