

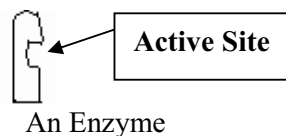
Yeast and Gas production

Planning (a)

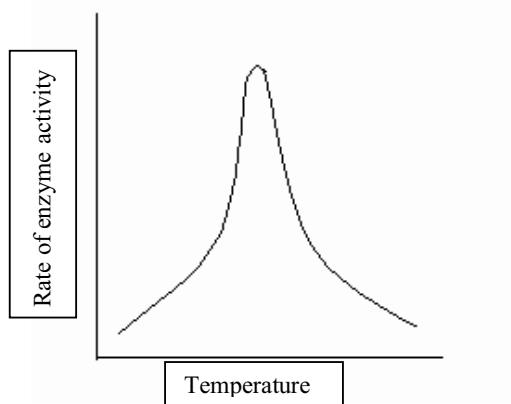
Aim: To investigate gas production by yeast in a glucose mixture.

Theory: The process by which living organisms release energy from their food is called *respiration*. It is the oxidation or break down of glucose molecules to release energy. Respiration can be of two types Aerobic and Anaerobic. During Aerobic respiration glucose is burnt in the presence of oxygen to produce energy and carbon dioxide. However in Anaerobic respiration glucose is burnt in the absence of oxygen to produce energy and alcohol or lactic acid (animals). Yeast cells respire anaerobically to produce ethanol and carbon dioxide. Yeast cells have enzymes which are used in this process, *Zymase* enzymes.

Enzymes are biological catalysts that alter the rate of reaction without being chemically changed during the reaction. They help to speed up the bio-chemical reactions in living organisms. Enzymes are substrate-specific. That is a particular type of enzyme only catalyses few reactions. This means that there are very less possible substrates that can combine with the active site of the enzymes. The active site of an enzyme is the region on the surface of an enzyme to which the substrates bind and are catalyzed by the enzyme. This active site has a particular shape and different chemical properties. Only the few possible substrates with the right shape and the chemical properties are attracted towards the active site and bind together to be catalyzed. This is often referred to as the **lock and key mechanism**.
the food substances.



A graph showing the rate of enzyme activity against temperature:



Enzymes speed up biological reactions by reducing the activation energy required and hence enabling the reaction to take place more readily.

The enzyme activity is dependant on temperature. At a low temperature the enzyme activity is slow, as the substrate molecules collide with the enzyme molecules less frequently. As the temperature increases the molecules vibrate faster and collide more frequently. This increases the rate of reaction as now more substrate molecules bind with the free active site and are catalyzed. However, as the temperature further increases $>60^{\circ}\text{C}$, the vibrations and the frequency of collisions increases, however the enzymes start to become denatured. At very high temperature the increases vibrations in the enzyme molecules break the bonds and the active site is lost. When this happens the enzyme is said to have **denatured** that is: lost its shape so that it can no longer perform its functions.

Hypothesis:

It is known that yeast respire anaerobically in a glucose mixture to produce carbon dioxide gas and alcohol (Ethanol- $\text{C}_6\text{H}_5\text{OH}$). This process is catalyzed by enzymes. Thus the rate of gas production will be the highest when the enzyme activity is maximum. The optimum temperature that is the temperature at which the enzymes work the fastest is around 40°C - 45°C . Therefore this is the temperature at which the rate of respiration will be the fastest. At a low temperature ($<20^{\circ}\text{C}$) the rate gas production will be slow as the enzyme activity is slow. At a very high temperature ($>60^{\circ}\text{C}$) the enzyme activity is again slow and hence the rate of gas production will also be slow.

Variables

Dependant variable: The dependant variable here is the rate of carbon dioxide production. As the temperature is varied the arte of carbon dioxide production and hence the rate of respiration is varied accordingly. If the temperature increases to about 40°C the enzyme activity increases and the carbon dioxide production also increases. However if the temperature is lowered to below 20°C or increased to above 60°C the enzyme activity decreases and thus the rate of gas production also decreases.

Independent variable: The independent variable here is the temperature. The temperature is varied to observe the changes in the rate of carbon dioxide production and hence in the arte of enzyme activity.

Controlled Variable: The controlled variable here is the Ethanol (C_6H_5OH) production. As the rate of respiration increases the rate of ethanol production also increases. However in this experiment it is important that this production is controlled. This is because Ethanol is a toxic substance and it stops the yeast growth. Therefore, excess of ethanol will decrease the yeast growth. The rate of respiration may go down and thus the rate gas production. This change will be due to the excess ethanol and not any variation in the temperature. Therefore it is important that the ethanol production is more or less kept constant so that it does not affect the rate of respiration as then there will be no limiting factor for the yeast cells. To do this we have to change the yeast and glucose solution after each reading to make sure that there is no initial toxic that will be inhibit the yeast growth.

Planning (b)

Apparatus:

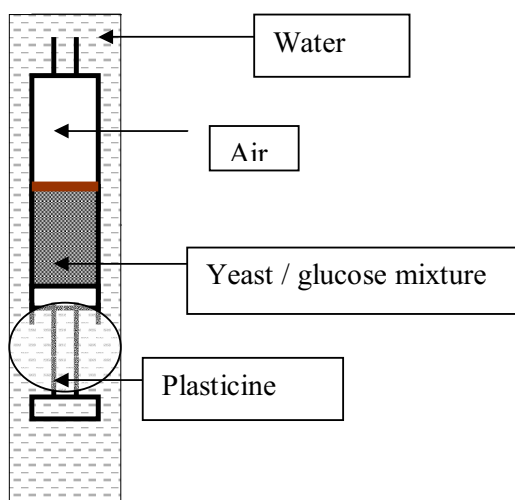
- Yeast in a glucose mixture
- Beakers
- Boiling test-tube
- Syringe 5ml
- Plasticine
- Test tube rack
- Thermometer
- Stop-watch

Procedure:

- First we fill a test tube with water at a particular temperature. (Varying from $20^{\circ}C$ - $65^{\circ}C$).
- Then we fill a syringe 2ml of yeast and glucose solution and then with 1ml of air.
- We then stick plasticine behind the syringe to make it heavy and completely be immersed into the water in the test tube.
- After about 2-3 minutes of the syringe being immersed in water the stop watch is started and the number of bubbles produced in 2minutes is counted and recorded.
- The experiment is repeated with water at different temperatures.

This is how the apparatus, which has been set up, will look like.





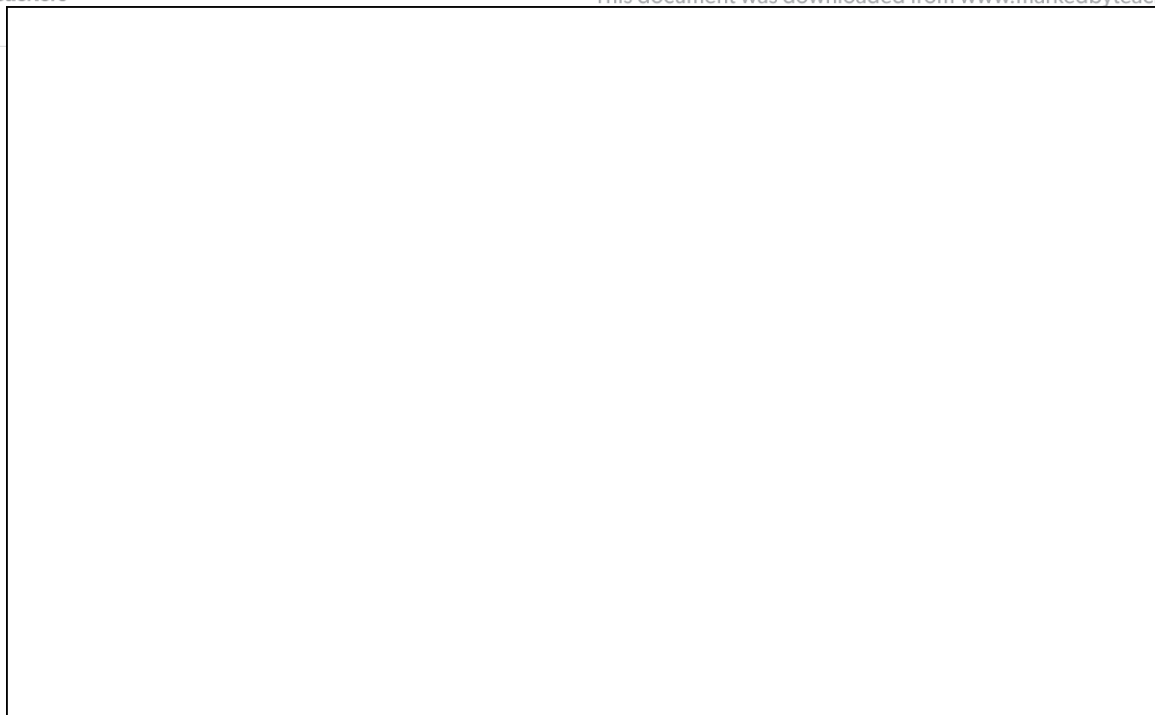
Data Collection:

It took some time for the bubbles to start coming out and that's why we are asked to start counting after 2 minutes. After this time bubbles came out at regular intervals for each given temperature. The readings of this experiment are given in the table followed. If you observed the test tube for even longer time, the number of bubbles produced fall after a particular time. This because the glucose solution was completely digested and there was no more of it left. So, the least and maximum temperatures gave bubbles for a longer period of time than the optimum temperatures as it took them more time to digest.

Sno.	Temperature/ $^{\circ}\text{C}$ $\pm 0.1^{\circ}\text{C}$	Time/seconds $\pm 0.01\text{s}$	No. of bubbles ± 1
1	20	120	0
2	35	120	3
3	45	120	5
4	60	120	3
5	65	120	2

Date analysis:

From the graph we can see that the maximum rate of gas production is at 45°C , thus the rate of reaction is maximum at this temperature.



Evaluation:

From the graph we see that at low temperature the rate of gas production or the rate of bubble production is zero. However as the temperature increases the rate of bubble production also increases and reaches a maximum at a temperature of 45°C. But in accordance with our hypothesis, as the temperature is further increases the rate of bubble production goes down. From this it can easily be concluded that the rate of gas production that is the rate of respiration of the yeast cells is catalyzed by *enzymes*. The enzyme activity is in turn dependant on the temperature and as the enzymes activity is affected by the temperature changes the rate of respiration is also changed.

In this experiment we took a syringe filled with approximately 2ml of yeast and glucose mixture and then filled with 1ml of air. This syringe was then inserted in the test

tube filled with water at a temperature at which we tested the production of gas. To obtain an accurate result a few things have to be kept in mind while doing the experiment:

- The glucose and yeast solution must be changed after at least two readings each in different temperature water. If this is not done the gas production may be slow not because of the temperature changes but due to the lack of glucose or due to the excess of toxic like ethanol which obstructs the growth of yeast cells.
- It must be ensured that before we start counting the bubbles the syringe is in the water for more than about 2 minutes so as to allow the solution to come to the same temperature as the water and the yeast cells to adjust to that temperature.

Besides these there are a few limitations to this experiment:

- The rate of gas production is measured by counting the rate of bubble production. However this is not very accurate as each time the size of the bubbles differ and hence volume of gas.
- Moreover, we know that the gas being produced is carbon dioxide which is soluble in water. Hence the accurate amount of gas production may not be known as some of it will dissolve in water.

Limitations

The main limitation of this experiment is that the temperature of the water surrounding the syringe will lose heat to its surroundings thus decreasing the temperature of the solution inside the syringe and sometimes might lead to give faulty readings. Another limitation could be the amount of yeast solution and glucose solution put into the test tube. This must not vary as it will cause more digestion and give more bubbles per minute for the temperature of water. Another limitation could be that by mistake we might count the bubbles that come out when water is shook, and this might give us another faulty count and thus our results could be proved wrong.

Precautions

Always make sure that the ratio of glucose solution to that of the yeast remains the same as this is very important and mistakes might give faulty readings. See to that you don't waste a lot of time after adding the yeast to the glucose other than waiting for two minutes. If you wait any longer the digestion will occur and it is possible that you might not see any bubbles when you actually start taking readings.

Modifications:

- Next time I would take a fresh glucose and yeast mixture after each reading so that the solution or the condition of the yeast cells is the same for all the temperature readings.
- Instead of counting the bubbles to measure the gas production I would collect the gas in a test tube and then measure its volume. This would give a more

accurate reading for the gas production as the error due to the size of the bubbles is overcome.

Conclusion

Through the experiment we were able to determine our aim and our hypothesis was also verified and proved right through our observations. We also were able to find the optimum temperature for the yeast and at what rate it digested at.