Enzyme Specificity Experiment.

Aim

To observe whether enzymes are specific and if so, which ones work best with yeast-therefore ferment.

Hypothesis

Sugar -> ethanol + carbon dioxide + energy
$$C_6H_{12}O_6$$
 -> $2C_2H_5OH$ + $2CO$ + energy

The sugar (substrate) used in this fermentation can vary. This experiment uses the amount of CO_2 evolved to determine which sugars yeast enzymes can use.

Materials

- Distilled water
- Glucose
- Galactose
- Fructose
- Sorbose
- Yeast

Apparatus

- Fermentation tubes x5
- Test Tubes x5
- Pipettes x5
- Temperature controlled water bath at 30°C
- Ruler

Diagram

Method

For the control, 10 drops of distilled water were placed in a fermentation tube along with 10 drops of yeast. This fermentation tube was then topped-up to the brim with more distilled water. The fermentation tube was then place upside down in water filled test tubes and the level of the fluid was recorded. After the level of the fluid was recorded and the other tubes completed they were put into the temperature controlled water bath. The procedure was the same for the other substrates - glucose, galactose, fructose and sorbose except that the original 10 drops of distilled water was replaced with the respective sugar. The tubes were left in the temperature controlled water bath for 40 minutes at 30°C. 1 hour was the original plan but because of time constraints was impossible to achieve.

Results

Substrate	Amount of displaced water (Cm ³)
Control (Distilled Water)	0.2
Glucose	2.9
Galactose	0.2
Fructose	2.7
Sorbose	0.5

Sources Of Error

- 1. An air bubble may have become trapped in the fermentation tube, this would add to the water displaced.
- Some of the liquid might have overflowed. With the fermentation tubes being full to the brim it is highly likely that some of the liquid will have spilt, therefore the amount of liquid in the tube would not be correct and may not be the same volume as all the other samples and this should influence the result.

- 3. The measurements for the liquids were not exact. We cannot be sure that the teat pipettes were measuring the same volume of liquid and a 'drop' may have been of different volume.
- 4. Initially the liquid in the tube was not at the same temperature as the water bath and so had to heat up so the time spent in the water bath is not equal to the time spent at 30°C
- 5. The liquids in the fermentation tubes may not have been mixed together at all or some may have been more than others, this means that the amount of liquid in contact with the yeast may have differed in the different fermentation tubes
- 6. The yeast may have already started to denature because of its age and so may not have given the correct results
- 7. As the water was expelled from the tube the water around it rose, if the water around the tube rose above the air bubble at the top of the tube it would have tried to flood back into the tube, compressing the gas. Similarly, at the start of the experiment the water pressure was stretching the air bubble because it was trying to get out to equalise the level of water inside the tube with that of the water outside the tube.
- 8. If the yeast was left in its container longer than a few minutes and not shaken before a sample taken-it will begin to settle and not a correct mixture of all components would have been collected.

Discussion

I think the enzymes act on the OH bond. The control sample was used to ensure that no gas was produced, and if any was the result from tube a was subtracted from the results of the other sample. This is because the result from tube a is the amount of gas produced when yeast reacts with distilled water-and as there is distilled water in all of the other samples, this is a factor that had to be taken into consideration.

Conclusion

The only sugars that worked well were glucose and fructose. The sorbose worked partly and the galactose didn't work at all, although from the results it showed some fermentation it is likely that it is just trapped air-as from the theoretical side of the experiment, it was known that galactose does not ferment with yeast. (See sources of error).