MICROSCALE CATALASE

A microscale enzyme study

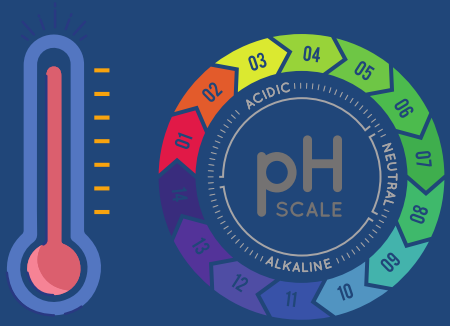
Catalase is a common enzyme found in nearly all living organisms exposed to oxygen. An enzyme is a protein that speeds up biological reactions. Catalase speeds up the break down of a harmful chemical called hydrogen peroxide, which is a by-product of cell metabolism. The products of this reaction are oxygen and water.

hydrogen peroxide (substrate) → catalase (enzyme) → **oxygen + water** (products)

Without catalase, hydrogen peroxide would cause oxidative damage and the production of reactive oxygen species. Consequently, catalase is crucial to protecting the cell from these potentially damaging species.

A number of factors are known to affect enzyme reactions, including:

- temperature
- pH
- substrate concentration



Each of these factors could be investigated using the protocol below.

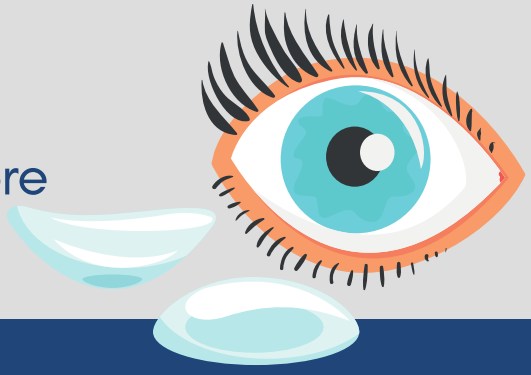
COMMERCIAL USES OF CATALASE

- In the food industry for removing hydrogen peroxide from milk prior to cheese production.

- In food wrappers where it prevents food from oxidising.

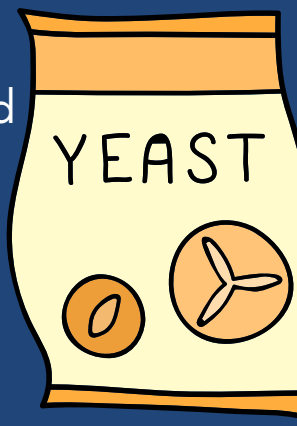
- In the textile industry to remove hydrogen peroxide from fabrics to make sure the material is peroxide-free.

- In contact lens hygiene – a few contact-lens cleaning products disinfect the lens using a hydrogen peroxide solution. A solution containing catalase is then used to decompose the hydrogen peroxide before the lens is used again.



HEALTH & SAFETY

When starting an experiment with a class, you must ensure you are comfortable and familiar with the risk assessment. What are the hazards, the level of risk and control measures that must be put in place? Is the current risk assessment appropriate for your learners? Do you need to make adjustments? This page on the SSERC website provides a Risk Assessment template and information about Dynamic Risk Assessments.



HAZARD	RISK	CONTROL MEASURES
Yeast suspension	Baker's yeast is a Level 1 organism, as outlined in the SSERC Code of Practice, and poses no risk to learners. Can be used in experiments by teachers with no specialist training.	Suspensions of Baker's yeast can be disposed of using good domestic hygiene practices. Page 7 of the Code of Practice provides full guidance, if required.
Hydrogen peroxide SSERC hazardous chemical database	This protocol suggests small volumes of 20vol hydrogen peroxide. At this concentration, a "health hazard" warning is highlighted: • risk of skin burns • eye damage • harmful if inhaled or swallowed	Wear eye protection. Teacher or technician should dilute the hydrogen peroxide to the working concentration(s). For **S1-S2 learners**, the concentration of hydrogen peroxide could be reduced to **1 vol, 5vol and 10vol**.

MATERIALS

This experiment investigates the effect of substrate concentration on enzyme activity. The substrate is hydrogen peroxide and the enzyme is catalase. As oxygen is formed, foam will be observed on the activity boards. The diameter of the foam circle should be measured, using a ruler, as a measure of enzyme activity.

AIM

To investigate the effect of substrate concentration on catalase activity.

INDEPENDENT VARIABLE

Concentration of hydrogen peroxide.

DEPENDENT VARIABLE

Catalase activity, measured indirectly by recording the diameter of the foam circle.

SAMPLE SIZE

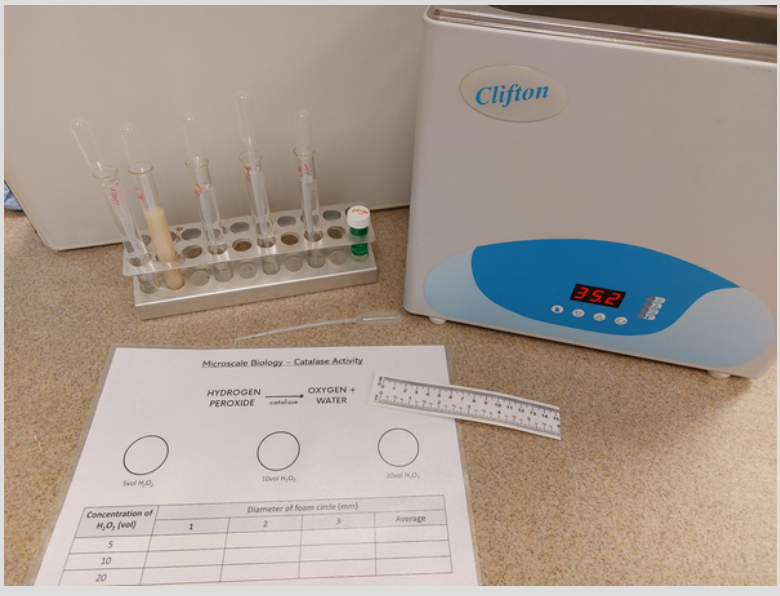
The experiment should be repeated three times at each substrate concentration. This should only be increased if the results are very variable.

SOURCE OF ENZYME

Baker's yeast suspension.

MATERIALS REQUIRED (PER PAIR)

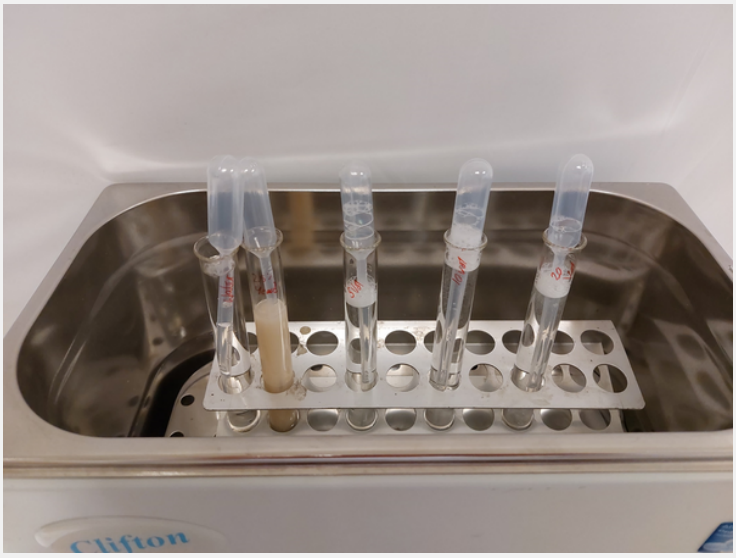
- 2.5% Baker's yeast suspension
- Test tubes containing:
  - 10ml 5vol hydrogen peroxide
  - 10ml 10vol hydrogen peroxide
  - 10ml 20vol hydrogen peroxide
- detergent
- distilled water
- 6x 3ml plastic pipettes
- ruler
- waterbath at 35°C
- stopwatch
- activity board.



METHOD

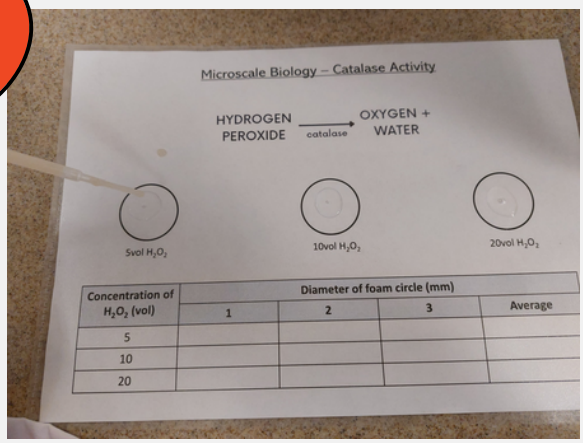
STEP 1

Add 2 drops of detergent to the test tubes of hydrogen peroxide, using a clean pipette. Put the test tubes of hydrogen peroxide and the yeast suspension into the waterbath to equilibrate to the optimum temperature (35°C).



STEP 2

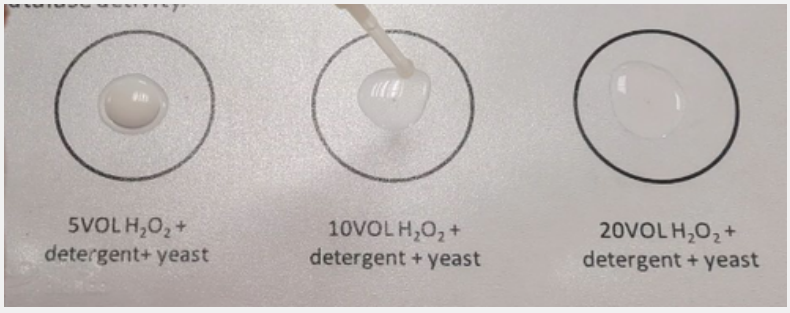
Use the table below to add the reagents to the circles on the activity board. Always add yeast last, as this is the source of the catalase enzyme.



Reagent	Drops of reagent to add to circle		
	5 vol	10 vol	20 vol
H_2O_2 + detergent	5	5	5
Water	3	3	3
2.5% yeast	2	2	2

STEP 3

Leave the reaction for 3 minutes and then measure the diameter (in mm) of the foam circle, using a ruler. Wipe the activity board clean and repeat the experiment a further two times.

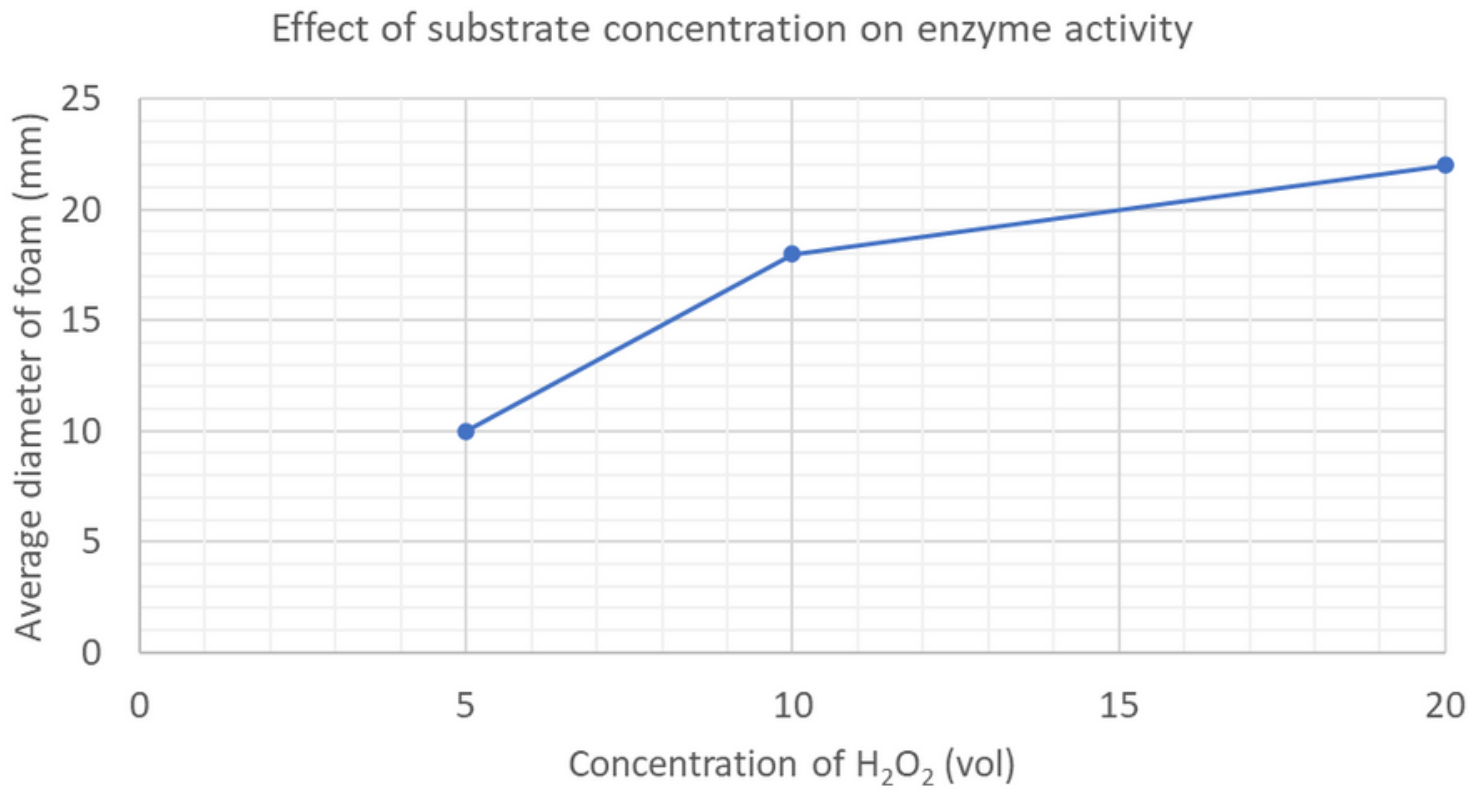


RESULTS

Concentration of H_2O_2 (vol)	Diameter of foam (mm)			
	1	2	3	Average
5				
10				
20				

TYPICAL RESULTS:

Concentration of H_2O_2 (vol)	Diameter of foam (mm)			
	1	2	3	Average
5	10	11	10	10
10	17	17	19	18
20	22	21	24	22



Learners should now reflect on their aim to form a conclusion. Another useful exercise at this point is to evaluate the method.