

AQA Biology GCSE

RP 05: Enzymes

Practical notes

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Effect of pH on the rate of activity of amylase enzyme

Aim

Use continuous sampling with iodine solution to determine the time taken for starch (the substrate) to be completely digested, hence calculate the rate of enzyme activity at different pH values.

Equipment

- test tubes
- a test tube rack
- water baths (electrical or Bunsen burners and beakers)
- spotting tiles
- a 5 cm³ measuring cylinder
- syringes
- a glass rod
- a stopwatch
- starch solution
- amylase solution
- buffered solutions
- iodine solution
- thermometer

Method

- 1. On a tile, label each well with the time (from 0 onwards) and add a drop of iodine solution to each well.
- 2. Add 2 cm³ of each buffer solution using a syringe (ranging from pH 3.0 to 7.0) into each labelled test tube.
- 3. Immerse the starch solution, amylase solution, and the test tubes of buffer solution in a water bath at 25°C.
- 4. Allow a few minutes for the temperature to equilibrate.
- 5. Use a syringe to add 2 cm³ of amylase into a test tube of buffer solution.
- 6. Use a syringe to add 2 cm³ of starch into the same test tube and start timing immediately.
- 7. Use the glass rod to transfer a drop of the mixture to the well labelled '0' on the tile.
- 8. Repeat step 6 every 30 seconds, rinsing the glass rod in between every test, until the iodine solution remains brown and does not turn blue-black.
- 9. Calculate the rate of enzyme reaction by using 1/ time taken for iodine solution to remain brown.
- 10. Repeat steps 2-8 for buffer solutions with different pH values.
- 11. Plot a graph of the rate of enzyme reaction against pH.

рН	Time taken for amylase to completely break down all the starch / s	Rate of reaction / s ⁻¹









Sources of error

Intervals between testing may be too long to accurately find the time taken for starch to be completely broken down.

Potential Hazards

Be careful using hot water.

If using a Bunsen burner tie long hair back and wear goggles.







