



## INVESTIGATING ENZYMES

Investigating the factors affecting enzymes experimentally

### How Science Works

A practical involving investigating the effect of determinants of enzymes is a favourable way of completing your Biology coursework section (the Practical Skills Assessment)

The basics of enzyme action are that enzymes are used to *catalyse* a reaction, where *substrate* is turned into *product*. The substrate occupies the *active site* of an enzyme

A practical involving enzymes will usually ask you to find the relationship between enzyme action (and rate of reaction) versus one of the determinants affecting the reaction rate – these may be temperature, enzyme concentration, substrate concentration, or pH – and you will have to perform an experiment to manually calculate this relationship

It is important that when carrying out the practical, all *other* variables except for the **dependent variable** and the **independent variable** are controlled. The dependent variable (DV) is the variable which is being studied, and the independent variable (IV) is the one which you, as the experimenter, are manipulating. So, for example, in a study to see the effect of temperature on rate of reaction, the DV is rate of reaction, and the IV is temperature, and all other variables (**extraneous variables**, or **control variables**) must be *controlled* – i.e. remain constant.

Below is a table which explains how each type of variable would be kept constant. In your experiment, you will *not* need to control all of these variables, as one will be your IV and another your DV. But the other ones from the table must be.

Variable	Method of control	Reason for method of control
Temperature	Carry out the reactions in a water bath with a thermostat	Room temperature is never constant and is too variable, fluctuations in the temperature will affect your results
Enzyme concentration	Use accurately-measured volumes of enzyme in solution	Reaction rate depends on the concentration of enzyme, so having this kept constant will lead to valid results
Substrate concentration	Use accurately-measured volumes or masses of substrate	Reaction rate depends on the concentration of substrate, so having this kept constant will lead to valid results
pH value	Use a pH buffer (a solution which maintains pH at a set level by keeping the hydrogen ions – proton donors – in the solution at a constant level)	The pH of the solution will have an effect on the active site of an enzyme, and so it is important that this is controlled for accurate results

You may be asked to provide reasons for keeping the above variables constant in your chosen experiment as part of the written accompaniment of the practical exam, so it is important that you understand the biological and scientific reasons behind each of the above methods of controlling the variables.

You will also no doubt need to calculate **reaction rate**. Reaction rate is most often calculated using the formula:

$$\text{reaction rate} = \frac{1}{\text{time}}$$

But it can be any variation of that, such as 10/time, or 100/time. Most likely you will be given the formula you're expected to use in the exam paper.

It is also important that you understand the need to carry out **repeats**, in order to obtain *reliable* results, and identify any **anomalous** results.